

# New Insights into the High-Pressure Processing of Meat and Meat Products

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**Abstract:** For years, high-pressure processing has been viewed as useful for pasteurizing food while maintaining the quality of fresh food. However, even at moderate pressure, this process is not without effects on food, especially on meat products. These effects are especially important because pressure greater than 400 MPa is generally necessary to achieve efficient microbial inactivation. In this review, recent advances in the understanding of the impacts of high pressure on the overall quality of raw and processed meat are discussed. Many factors, including meat product formulation and processing parameters, can influence the efficiency of high pressure in pasteurizing meat products. It appears that new strategies are applied either (i) to improve the microbial inactivation that results from high pressure while minimizing the adverse effects of high pressure on meat quality or (ii) to take advantage of changes in meat attributes under high pressure. Most of the time, multiple preservation factors or techniques are combined to produce safe, stable, and high-quality food products. Among the new applications of high-pressure techniques for meat and meat-derivative products are their use in combination with temperature manipulation to texturize and pasteurize new meat products simultaneously.

## Introduction

High-pressure processing is a technology by which a product is statically treated at or above 100 MPa by means of a liquid transmitter. High pressure, also called high isostatic pressure, has traditionally been used in the production of ceramics, steel, superalloys, and synthetic materials. The fact that high pressure kills microorganisms and preserves food was discovered in 1899 by Bert H. Hite (1899). However, this technology has only been thoroughly investigated in biological and food systems since the early 1980s, and the first pressure-treated product (jam) reached the Japanese food market in 1990 (Knorr 1993).

High-pressure processing has various advantages over other non-thermal technologies used to improve food safety. Food can be processed at ambient or even lower temperatures. Due to the isostatic transmission of pressure, the processed material experiences the pressure instantaneously with no gradient, resulting in uniform treatment irrespective of the size and geometry of the material. High-pressure modifies only noncovalent bonds and does not affect small molecules such as flavor compounds and vitamins; therefore, high-pressure processing leads to less degradation in the overall quality of processed foods in comparison with heat-treated foods.

As a result, high-pressure processing enables food manufacturers to respond to the growing demand for safe, fresh-looking, nutritious, and innovative food products. Recent progress in

equipment design has improved access to high-pressure devices. Safe, readily cleanable, automated, and mass-produced equipment made of stainless steel is currently available (Tonello 2008). Consequently, the use of high-pressure technology in food processing has steadily increased over the past 10 yr. Among products processed using high pressure, the number and variety of meat and meat products has risen dramatically worldwide (Garriga and Aymerich 2009). Products are mainly processed with high pressure to increase their safety by inactivating microorganisms (mostly *Listeria*) without altering attributes contributing to sensory quality. Such high-pressure-treated products are mostly found in the United States and Japan. In Europe, Spain is a pioneer in high-pressure-treated meat and first commercialized sliced cooked ham in 1998. The ham was treated at 400 MPa and 17 °C for 20 min after vacuum-packaging and could be stored for 8 wk at 4 °C (Grebol 2002). Cured ham and some precooked meals containing poultry, pork, chorizo, and various types of sausages are now available on the Spanish market (Garriga and Aymerich 2009).

However, although the changes induced in food by the use of pressure are different from those occurring in foods that are processed using heat, these changes are not negligible. Indeed, such changes are variable and respond to the Le Châtelier principle, meaning that reactions accompanied by a decrease in volume are enhanced by pressure. Pressure affects the conformation of macromolecules, the transition temperature of lipids and water, and a number of chemical reactions (Tauscher 1995). The effects may be beneficial or detrimental depending on various factors, such as processing parameters or product formulation (Rastogi and others 2007). Preserving meat quality and controlling pressure-induced changes in meat products are important issues for meat-product manufacturers.

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Meat quality is a highly subjective topic, but industry and consumers agree on a number of important quality indicators. These traits include tenderness, juiciness, appearance (color and structure), fat and protein content, drip and cooking loss, fat quality (namely the oxidative stability of fat), and off-odors (Borggaard and Andersen 2004). The effects of high-pressure treatments on meat and meat-product quality have been extensively studied over the past 40 yr and were reviewed by Cheftel and Culioli (1997). The current review focuses mainly on work and insights from the last 15 yr. Early studies clearly showed that significant and irreversible modifications occur in meat as a result of high-pressure treatments. These changes continue to be studied, and numerous recent publications focus on oxidation and microstructure.

The objectives of this review are, first, to present recent knowledge regarding the effects of high-pressure treatment on raw meat and meat products and, second, to identify the limitations and potential of high-pressure treatments for meat and meat products. For this purpose, the review is divided into four parts. First, recently acquired fundamental knowledge about the effects of high pressure on meat and meat-product quality is presented. High-pressure effects are very variable, and the literature is analyzed to highlight the main factors affecting this variability. Because high pressure is most often used to pasteurize food products, the second part of the paper addresses microbial inactivation in meat and meat products. Inactivation is also highly variable; hence, the main factors affecting it are analyzed. One important challenge associated with high-pressure technology is ensuring high levels of microbial inactivation in meat products while maintaining those sensory characteristics that ensure their fresh appearance. For this reason, most recent studies have addressed strategies to improve the safety of meat products while maintaining good sensory quality attributes either by combining high pressure with other technologies or by modifying product formulations. These studies are reviewed in the third part of the paper. Finally, the fourth part examines new opportunities for high-pressure treatments of meat and meat products that are distinct from pasteurization.

### Recent Advances in Knowledge of the Effects of High-Pressure Treatment on the Quality Attributes of Meat and Meat Products

In the following discussions, temperature is not specified when treatment was performed at ambient temperature.

#### High-pressure effects on raw meat tenderization and texture

Since the 1980s, many studies have been performed to understand pressure effects on meat quality. Meat texture was the most investigated quality attribute in early works because of interest in using high pressure for meat tenderization. It is well known that tenderization of fresh meat occurs due to the following changes in the muscle during conditioning (mainly as a result of the activity of endogenous proteases): weakening of actin–myosin interactions, fragmentation of myofibrils into short segments as a result of Z-line disintegration, degradation of elastic filaments consisting of connectin, and the weakening of connective tissue (Koochmarai 1994). The study of the effects of high pressure on meat proteins (enzymes and myofibrillar proteins) partly explains the modification of the texture and tenderness of raw meat that occurs under pressure.

Early work has shown that pre-rigor high-pressure treatments of muscles at approximately 100 MPa and 30 °C generally lead to a substantial shortening of the muscle (approximately 35%) and

an improvement in tenderness after cooking (Macfarlane 1973; Bouton and others 1977). Such a degree of shortening (35%) would be expected to result in considerable toughening (Locker and others 1960; Davey and others 1967). The improved tenderness has been suggested to be linked to the effect of pressure on the contraction state of the muscle (Macfarlane 1973). In fact, the physical disruption of sarcoplasmic reticulum membranes under pressure leads to an increase in cytosolic  $\text{Ca}^{2+}$  (Okamoto and others 1995). The release of  $\text{Ca}^{2+}$  results in intense muscle contraction, an acceleration of postmortem glycolysis and a rapid pH decrease (Macfarlane 1973) due to activation of  $\text{Ca}^{2+}$ -dependent phosphorylases involved in the regulation of glycogen breakdown (Horgan and Kuypers 1983). The combining effects of muscle contraction and pressure during the treatment could lead to breakage of myofibrillar structure, forcing myosin filaments of severely contracted muscles into Z discs, which would explain the tenderizing effect (Macfarlane 1973).

Tenderization has also been attributed to the activation of two enzymatic systems involved in the tenderization of meat during aging, namely cathepsins and calpains. Cathepsins are released from lysosomes when the muscle is high-pressure treated at 100 MPa just after animal death, and they can be absorbed rapidly by the myofibrils (Kubo and others 2002). Calpain activity in pressure-treated muscle is increased by pressure up to 200 MPa due to the activation of the calpain proteinase system by  $\text{Ca}^{2+}$  released from the sarcoplasmic reticulum and to the inactivation of the inhibitor calpastatin under pressure (Homma and others 1996). However, the influence of high pressure on calpain system activity has been shown to be complex: *in vitro* calpain activity was both favored for moderate pressure (50 MPa) and inhibited at higher pressure levels ( $\geq 100$  MPa) (Bessiere and others 1999).

Some researchers have also shown interest in the high-pressure activation of a cytosolic proteinase complex (a proteasome) that has been isolated from a wide variety of eukaryotic cells and tissues (Rivett 1993). The proteasome isolated from muscle is able to degrade myofibrillar proteins and is also presumed to be involved in the degradation of muscle proteins during postmortem storage (Otsuka and others 1998). *In vitro* analysis has demonstrated that high pressure (<200 MPa) could activate this proteasome (Otsuka and others 1998; Yamamoto and others 2005).

Recent investigations have focused on the application of high pressure to post-rigor muscles because pre-rigor treatment involves a hot-boning treatment that is impractical in those plants that practice traditional cold-boning. As is the case for pre-rigor meat, it has been reported that pressure induces the release of cathepsins from lysosomes in post-rigor muscles and provides a gradual increase in activity with increasing pressure up to 400 MPa (Homma and others 1994). Jung and others (2000b) also observed an increase in cathepsin D activity related to the breakdown of lysosomal membranes in post-rigor beef muscle treated at 520 MPa for 260 s in comparison with untreated muscle. In addition, Jung and others (2002) showed that such a high-pressure treatment does not disturb the recognition between cathepsin D and myofibrils *in vitro*.

The activity of all enzymatic systems involved in meat tenderization may also be modulated by pH variation. Generally, pH decreases reversibly during a pressure treatment due to the changing dissociation constants of attendant acids and bases (Stippel and others 2004), and the pH of post-rigor meat increases slightly (by approximately 0.5 pH units) immediately after the pressure treatment (Hugas and others 2002; Sikes and others 2010). These pH changes may also contribute to the influence of pressure on

enzymatic activity in meat; for example, the drop in pH under pressure could contribute to the activation of cathepsins, which have optimal pH below 5.2 (Faustman 1994).

However, although some key enzymes in the tenderization process can be activated by high pressure at or below room temperature in post-rigor meat, this activation is not necessarily accompanied by a decrease in meat toughness as observed in pre-rigor meat. In the work of Jung and others (2000b), the activation of catheptic activity at 560 MPa had no conclusive effect on the tenderness of meat; instead, an increase in the shear force was observed in comparison with nontreated samples. Meat hardness also increased after high-pressure treatment at or above 200 MPa at 20 °C in beef muscle (Ma and Ledward 2004) and turkey breast fillets (Del Olmo and others 2010). No improvement in tenderness was observed after cooking high-pressure-treated beef muscle at 70 °C in comparison with an untreated cooked sample (Ma and Ledward 2004), in agreement with earlier reports (Macfarlane 1973; Bouton and others 1977). Only two recent studies report a tenderizing effect of high pressure at room temperature or lower in post-rigor meat. Suzuki and others (1992) and Schenkova and others (2007) reported that a decrease in the hardness of cow muscle could be obtained after a high-pressure treatment consisting of a brief exposure to pressures of between 100 and 300 MPa at 10 °C. It is not clear why these results differ from those previously reported, but the difference may result from the different methods used to evaluate meat tenderness. Suzuki and others (1992) used a conical plunger to evaluate raw meat hardness; Schenkova and others (2007) do not mention cooking before Warner-Bratzler shear force measurements. In all other mentioned studies addressing beef, Warner-Bratzler shear force measurements were performed on cooked meat. This difference in results could also be caused by variability in the aging of the high-pressure-treated meat.

When an increase in meat hardness or shear value was observed after a high-pressure treatment at ambient temperature or lower, these changes were linked to pressure-induced structural modifications of myofibrils (Jung and others 2000a; Scheibenzuber and others 2002). Irreversible structural modifications increase with the pressure level, and the level of pressure at which the first modifications are observed depends on the animal species. Jung and others (2000a) observed no structural change in beef meat myofibrils treated at 130 MPa at 10 °C, but high-pressure treatments at 325 and 520 MPa caused increasing modifications in the ultrastructure of myofibrils. In chicken and pork, myofibrils treated at 100 MPa at room temperature looked thicker than untreated samples, and morphological changes become obvious with higher pressure (Iwasaki and others 2006). The degree of myofibril fragmentation is increased by high pressure, as with aging, due to the dissociation of myosin filaments and the release of  $\alpha$ -actinin (Iwasaki and others 2006). Furthermore, other modifications occur in high-pressure-treated meat proteins, which could explain the increase in hardness. In turkey meat, a swelling of acto-myosin is observed for pressures of up to 300 MPa; this swelling then leads to an increase of meat hardness (Scheibenzuber and others 2002). At pressures higher than 300 MPa, myofibrillar cross-sections are completely modified: myofibrils appear as indistinguishable shapes and the myofilaments are dissociated (Jung and others 2000a; Scheibenzuber and others 2002). The coagulation and aggregation of certain sarcoplasmic proteins in inter-myofibrillar spaces can interfere with myofibril sliding and consequently increase meat resistance (Jung and others 2000a; Scheibenzuber and others 2002). The apparent high elasticity of the myofibrillar gel that is pressure-

treated at 100 to 300 MPa prior to heating (Iwasaki and others 2006) corroborates the fact that modifications of pressure-treated proteins (swelling, aggregation, gelation) are linked to an increase in hardness in whole meat.

Finally, long exposure to high pressures at moderate temperatures has been shown to be necessary to improve the meat tenderness of post-rigor muscles. In beef muscle, exposure for 20 to 30 min to 150 to 200 MPa at 60 °C is necessary to improve tenderness (Macfarlane 1973; Bouton and others 1977; Locker and Wild 1984; Ma and Ledward 2004; Sikes and others 2010). According to Ma and Ledward (2004) and Sikes and others (2010), the tenderness improvement could be due to increased protease activity at 50 to 60 °C under pressure (in comparison with high-pressure treatments at lower temperatures). Ma and Ledward (2004) also hypothesized that some loss of the collagen components of toughness may occur under these conditions. In fact, meat tenderization can sometimes be limited by so-called "background toughness" due to the presence of connective tissue and other stromal proteins (Ratcliff and others 1977). Changes in connective tissue during aging are minor in comparison with those that occur in the myofibrillar proteins, so it would be interesting to increase the tenderness of meat containing large amounts of connective tissue. Recent experiments have aimed to clarify the effects of high pressure on connective tissue. Like aging, high pressure (150 to 500 MPa for 5 min at 8 °C) can induce structural weakening of isolated intramuscular connective tissue, as revealed by histology (Ichinoseki and others 2006). However, Fernández-Martín and others (2000a) reported that, in raw pork and beef, connective proteins remain practically unaltered by treatment at a pressure of 200 MPa. In poultry meat treated at 400 MPa, collagen also remains thermally unaltered during pressure treatment at any temperature from 10 to 75 °C (Fernández-Martín 2007). Furthermore, the extractability of proteoglycan is unchanged in high-pressure-treated muscle (100 to 400 MPa for 5 min at 4 °C), contrary to aging (Ueno and others 1999). It thus seems that high pressure has no effect on connective tissue in whole meat.

High pressure has been shown to improve meat tenderness when applied to pre-rigor meat. In post-rigor meat, tenderization can be only achieved after long (20 to 30 min) exposure to 150 to 200 MPa at 60 °C. It is believed to be due to the activation of some proteases involved in meat aging. In general, high-pressure treatments performed at ambient temperatures do not improve the tenderness of post-rigor meat because of coagulation, aggregation, or gelation of sarcoplasmic proteins and myofibrils.

### High-pressure effects on water retention and texture of meat products

Meat products are defined as those in which fresh meat has been modified by any of several processing methods, including curing, comminution, dehydration, fermentation, or cooking.

The cooking of meat batters either before or after high-pressure treatment results in varying effects on meat product texture. High-pressure treatment of cooked sausages at 500 MPa and 65 °C gives less firm, more cohesive products with lower weight loss compared to heat-pasteurized sausages cooked at 80 to 85 °C for 40 min (Mor-Mur and Yuste 2003). High-pressure treatment of meat batters before cooking gives rise to more elastic gels than those found in cooked-only batters (Iwasaki and others 2006) and to reduced cooking losses (Sikes and others 2009). By simultaneously combining high pressure with thermal treatments (300 to 700 MPa, 40 to 60 °C), the gel strength of raw ostrich sausages increased, and the amount of released and expressible water

decreased with increasing severity of the treatment (Supavitpatana and Apichartsrangkoon 2007; Chattong and Apichartsrangkoon 2009). According to these authors, the increased concentration of solubilized myofibrillar proteins leads to greater water binding and fat emulsification of the resulting meat gels.

Combining high pressure and heat treatment is particularly interesting at meat protein denaturation temperatures (above 60 °C). For example, high-pressure treatment of meat batters at 200 or 400 MPa and at 70 to 80 °C leads to better fat and water retention than is found in cooked-only samples (Jiménez-Colmenero and others 1998a). If high pressure (200, 400, or 500 MPa) is applied at temperatures above 70 °C, the resulting texture of meat batters is different from that of heated-only batters and is characterized by lower hardness and apparent elasticity (Fernández-Martín and others 1997) but higher cohesiveness (Yuste and others 1999). At such denaturing temperatures, protein denaturation can be shifted toward higher temperatures under pressure compared to heated-only samples (Fernández-Martín and others 1997; Jiménez-Colmenero and others 1998b; Carballo and others 2000; Fernández-Martín and others 2002; Fernández-Martín 2007); which explains differences in the functional properties between pressure-heated and heated-only products.

Product composition can also greatly influence the texture and water retention of meat batters after a high-pressure treatment. High-pressure treatments (100 to 400 MPa at 10 to 20 °C) and salt (1 to 2%) interact to reduce the cooking loss of meat batters (Iwasaki and others 2006; Sikes and others 2009). This may be because increasing sodium chloride causes increasing denaturation of proteins in high-pressure-treated meat batters (Fernández-Martín and others 2000b) and favors the solubilization of proteins and the formation of a gel network that retains water (Iwasaki and others 2006; Sikes and others 2009). Moreover, there is a synergistic effect of tripolyphosphate and high pressure (400 MPa for 30 min at 70 °C) on water-holding capacity in pressure- and heat-treated pork batters (Fernández-Martín and others 2002), indicating that high pressure could compensate for a decrease in tripolyphosphate content.

The extent of water and fat release after high-pressure treatment of patties has also been shown to be influenced by fat content. High fat content (20% w/w) was associated with higher fat and lower water release on centrifugation, expressed as percent sample weight, of pressure-treated beef patties (300 MPa, 5 °C, 5 to 20 min) in comparison with low-fat (9% w/w) patties treated in the same way and with untreated high-fat patties (Carballo and others 1997). Whatever the fat content, fat release was higher in high-pressure-treated samples in comparison with untreated samples and could reach 4%, whereas it did not exceed 2.6% in untreated samples (Carballo and others 1997). A similar trend is reported by Jiménez-Colmenero and others (1997) for fat release by centrifugation of high-pressure treated (300 MPa at 6 to 8 °C for 5 to 20 min) low-fat (9% w/w) and high-fat (25% w/w) pork sausages. Indeed, Carballo and others (1997) found that high-pressure treatment favored the rupture of adipocytes in beef patties, leading to higher fat release. Moreover, these authors showed that the texture after cooking was independent of fat content in pressure-treated samples (Carballo and others 1997), whereas an increase in fat content would normally lead to a lower Kramer shear force in cooked-only samples. However, the addition of 1% hydrocolloids, such as carboxymethylcellulose, locust bean gum, or xanthan gum, can improve the distribution of fat in high-pressure-treated sausages at 600 MPa for 40 min at 50 °C, giving rise to less elastic-like behavior in comparison

with sausages with no hydrocolloids (Chattong and others 2007). Finally, the addition to meat products of ingredients that may gel under high pressure will affect the resulting texture of the product. Experiments have been performed with dried egg white (Trespacios and Pla 2009), starch (Fernandez and others 1998; Fernández-Martín and others 2000c), and carrageenan (Fernandez and others 1998). These ingredients generally increase the hardness and improve the binding properties of high-pressure-treated meat batters (Fernandez and others 1998; Trespacios and Pla 2009).

High pressure can also be used to create meat-to-meat binding in cold-set restructured meat products (Hong and others 2008a, b). Meat restructuring involves the use of specific ingredients or additives, among which NaCl plays an important role. The binding properties of 1-cm pork-meat cubes mixed with NaCl, tripolyphosphate, glucono- $\delta$ -lactone, and  $\kappa$ -carrageenan increased with high-pressure treatment at 200 MPa for 30 min at 4 °C due to the creation of a continuous network of  $\kappa$ -carrageenan in the junctions between meat cubes (Hong and others 2008a). In such restructured meat, the addition of glucono- $\delta$ -lactone has been shown to compensate for the reduction in salt because it produces acid-induced gelation under pressure (Hong and others 2008b). Meat binding with transglutaminase, an enzyme that catalyzes the cross-linking of several proteins (including beef and poultry actomyosin) via the formation of covalent nondisulfide cross-links, can also be enhanced under pressure. High pressure (200 to 300 MPa for 15 min at 40 °C) renders the protein substrate more accessible to transglutaminase in turkey breast muscle paste (Ashie and Lanier 1999), thereby enhancing intermolecular cross-link formation and gel strength. Microbial transglutaminase has been shown to be stable at pressures up to 400 MPa at room temperature (Menendez and others 2006). For these reasons, a combination of high pressure and use of transglutaminase in meat products with specific formulations offers new possibilities for the creation of products with improved texture (Trespacios and Pla 2007a, b).

In whole meat products, drip loss has sometimes been shown to increase with a high-pressure treatment in comparison with untreated products in some occasions. This is the case for cooked pork ham treated at 600 MPa and 20 °C for 10 min 48 h after the ham production for which Pietrzak and others (2007) found 9.1% drip loss after 8 d of chilled storage versus 7.4% for the control. On the contrary, after treatment at 400 MPa and 7 °C for 20 min, no difference in drip loss was observed in cooked ham in comparison with untreated cooked ham (López-Caballero and others 1999). These different results for drip loss could be due either to the different temperatures or to the different pressure levels. According to Korzeniowski and others (1999), the amounts of drip loss and free water depend on the pressure level applied to pork meat at ambient temperatures. These factors increase with pressure treatment at 100 to 200 MPa (in comparison with untreated samples) and decrease at higher pressures (300 to 400 MPa) in comparison with samples treated at lower pressures. In this study, drip losses were 4% in untreated pork meat, reached 7% in samples treated at 200 MPa and decreased to 4% in samples treated at 400 MP. Moreover, the discharge resulting from heating samples that were previously high-pressure-treated at pressure levels of 300 to 400 MPa was significantly lower than that found for heated-only samples; however, if high pressure treatment was performed at 100 to 200 MPa, no difference was observed between heated-only samples and high-pressure-treated, heated samples.

The resulting texture and water binding of meat products can be explained by the protein gelation process. Various studies have



shown that high pressure causes depolymerization, solubilization, denaturation, and aggregation in myofibrillar proteins (Chefftel and Culioli 1997; Chapleau and others 2004). First, pressure-induced myosin denaturation leads to very different gels than those produced by heat; pressure (200 to 800 MPa for 20 min at ambient temperature) forms mainly heat-labile hydrogen-bonded structures, whereas heat (40 to 80 °C for 20 min) gives rise to structures that are stabilized by disulfide bonds and hydrophobic interactions (Angsupanich and others 1999). According to these authors, this is why pressure-treated muscles (400 MPa) are harder than samples that are cooked (50 °C), cooked and then pressure-treated, or pressure-treated and then cooked samples. Second, the mechanism of protein denaturation differs according to the pressure/temperature combination (Jiménez-Colmenero 2002; Lee and others 2007) in such a way that both the levels of pressure and temperature and the sequence in which they are applied are important. For example, pressure treatment (100 to 200 MPa for 20 min at ambient temperature) prior to heating (70 °C for 20 min) enhanced heat-induced gelation of myofibrils, leading to an improvement of gel elasticity (Iwasaki and others 2006), whereas heating under high pressure (200 MPa for 30 min at 70 °C) limits the gelling process of the meat system in comparison with heated-only meat batter (Jiménez-Colmenero and others 1998b). Finally, using nuclear magnetic resonance imaging (MRI), Bertram and others (2004) revealed that the interactions between muscle proteins and water are different in cooked beef that had been previously treated with pressure and heat (150 MPa, 60 °C, 30 min) compared with untreated cooked beef. According to Bertram and others (2004), these modified water properties can be linked to the lower juiciness that has been demonstrated with sensory analysis for beef treated with pressure and heat (150 MPa, 60 °C, 30 min) before cooking in comparison with cooked-only meat (Ratcliff and others 1977).

The texture of processed meat is also dependent on proteolytic activity in the product (Jiménez-Colmenero and others 1998b), which is partially linked to protease activity. In some processed meat, such as dry-cured products, enzyme activity is essential for the development of flavor components and their precursors. Particularly, cathepsin activity may affect the flavor and texture of products. As in raw meat, cathepsins have been shown to be highly pressure-resistant in processed products. Campus and others (2008) observed no effect of high pressure on the cathepsin activity of dry-cured pork loins and reported only a 20% decrease in activity after 45 d of vacuum storage at 4 °C in meat products treated at 400 MPa for 10 min at 20 °C. Furthermore, no effect of pressure was observed on cathepsin activity in frozen ham treated at 400 or 600 MPa for 10 min at 4 °C; the cathepsin activities were similar if the hams were frozen and high-pressure-treated at early processing stages of the dry-cured ham or at the end of the resting stage (Serra and others 2007b).

High pressure induces meat protein modifications (gelation, solubilization, aggregation) differently than heat-induced changes. High pressure results in varying effects on meat product texture and water retention, depending mainly on the product's form and composition, the pressure level, and the pressure/temperature combination. Combining high pressure and heat can sometimes improve the water retention in meat batters, depending on the batter's composition and the process parameters. High pressure can also be used to improve meat binding in restructured meat. Nevertheless, each specific product should be tested individually for the resulting effects of high pressure on its quality.

## High pressure's effects on oxidation of meat and meat products

When thermal treatment is applied to meat, lipid oxidation is the major cause of deterioration during subsequent storage. This is particularly the case for meats like chicken that contain significant amounts of unsaturated fatty acids. Lipid oxidation leads to rancidity and off-flavors, and a quantitative description of lipid oxidation in high-pressure-treated meat and meat products is needed for successful implementation of high-pressure technology in the meat industry.

A number of recent studies have addressed effects of high-pressure treatment on lipid oxidation in poultry meat and meat products. In most of these studies, oxidation was evaluated by measuring secondary lipid oxidation products, such as thiobarbituric acid-reactive substances (TBARS) or hexanal content. In several studies, oxidation was not increased immediately after the pressure treatment, but pressure induced lipid oxidation during subsequent storage of the meat (Dissing and others 1997; Orlien and others 2000; Beltran and others 2003, 2004a). However, Tuboly and others (2003) observed an increase in oxidation just after the pressure treatment. These authors reported the appearance of cholesterol oxidation products in high-pressure-treated turkey meat (400 MPa for 20 min) that was thawed for the treatment and refrozen for storage. The content of oxidation products increased immediately after the treatment, and after 4 and 8 mo of frozen storage, the content was higher in comparison with untreated meat stored under the same conditions.

The pressure-induced oxidation in raw poultry meat varies with the pressure level and, to a lesser extent, with the treatment time (Dissing and others 1997; Orlien and others 2000). Orlien and others (2000) treated vacuum-packaged chicken breast muscle at 300 to 800 MPa for 5 to 10 min at 25 °C. The authors reported an effect on lipid oxidation after 10 d of subsequent chilled storage for pressures above 600 MPa in comparison with untreated samples stored under the same conditions. The authors also showed that 800 MPa of pressure for 10 min was the most damaging pressure treatment with regard to lipid oxidation; for the production of secondary lipid oxidation products during subsequent chilled storage, it was equivalent to a heat treatment at 80 °C for 10 min. In the study by Dissing and others (1997), a pressure treatment of vacuum-packed turkey thigh muscle at 500 MPa for 30 min at 10 °C was equivalent to a heat treatment at 100 °C for 10 min in terms of TBARS found after 6 d of storage at 5 °C. Interestingly, for raw minced chicken thighs treated at 500 MPa and 50 °C for 30 or 60 min, vacuum-packaging prevented oxidation during 9 d of subsequent storage at 4 °C (Beltran and others 2004a). Nevertheless, in the latter study, oxidation was shown to significantly increase after 6 d if samples were stored in contact with air. Thus, vacuum-packaging could help in delaying oxidation of pressure-treated chicken meat, but the post-slaughter history and variations in the quality of the raw material may have different effects on the development of lipid oxidation after a high-pressure treatment. Ma and others (2007) compared oxidation in beef and poultry muscles after high-pressure treatments (0.1 to 800 MPa) at different temperatures (20 to 70 °C) and showed that chicken was more stable with regard to oxidation after pressure treatment than was beef. This result conflicts with the observations of Schindler and others (2010), who reported that, when treated at 400 or 600 MPa for 15 min at 5 °C, beef is more stable with regard to lipid oxidation than is chicken; this result should be expected given the smaller fraction of unsaturated fatty acids in beef

compared with chicken. In raw minced pork meat packed with air and pressure-treated at 200 to 800 MPa for 20 min at ambient temperature, Cheah and Ledward (1996) found that lipid oxidation appeared at treatments above 300 MPa after 1 d of chilled storage and that treatment at 800 MPa was equivalent to heat treatment at 80 °C for 15 min with regard to lipid oxidation.

Oxidation is also accelerated in ready-to-eat meat products. High-pressure treatment (500 MPa for 30 min at 50 °C) of minced chicken thighs increased oxidation during subsequent storage in comparison with cooked-only samples (50 °C for 30 min) (Beltran and others 2004a). In dry-cured ham, high-pressure treatment below 800 MPa (15 min, 20 °C) had little effect on lipid oxidation just after high-pressure treatment (Andrés and others 2004). In the study by Fuentes and others (2010), after one month of storage at 4 °C, high-pressure treatment had significantly enhanced the formation of lipid-derived aldehydes in dry-cured hams that were treated at 600 MPa and 12 °C for 6 min (in comparison with untreated ham stored under the same conditions).

The composition and physical treatment of meat products can greatly influence oxidation after high-pressure treatment. When added to chicken slurries, salt (5% w/w) was shown to have a pro-oxidant effect that was stronger for samples that were pressure-treated at 500 MPa for 30 min at 20 °C than for samples cooked in a water bath at 90 °C for 15 min (Beltran and others 2003). Moreover, the mechanical processing of meat (slicing or mincing) before a high-pressure treatment has a strong prooxidant effect (Beltran and others 2003; Fuentes and others 2010). However, mechanical processing after pressure treatment does not enhance oxidation (Beltran and others 2003). To minimize rancidity in pressure-treated meat products, several authors have proposed adding an antioxidant to product formulations. Rosemary and sage extract were found to be effective in retarding lipid oxidation during chilled storage in comminuted chicken treated with high pressure between 300 and 800 MPa at 10 to 20 °C for 10 to 30 min (Beltran and others 2004b; Bragagnolo and others 2007; Mariutti and others 2008). Rosemary was also found to protect tocopherols against degradation in chicken meat processed at 600 MPa and 10 °C for 10 min (Bragagnolo and others 2005). Other antioxidants, such as EDTA and egg white powder, have been shown to protect pressure-processed (300 and 500 MPa) chicken meat slurries against lipid oxidation during subsequent chilled storage due to their abilities to chelate metal ions (Beltran and others 2004b). Tume and others (2010) enriched beef muscle with  $\alpha$ -tocopherol by dietary means but failed to increase the lipid stability of the muscle under high pressure (800 MPa for 20 min at 60 °C) during 6 d of chilled storage. Finally, oxygen should be eliminated in high-pressure-treated meat packaging to avoid lipid oxidation during storage. This can be achieved either with a modified atmosphere with N<sub>2</sub> and CO<sub>2</sub> (Andrés and others 2006) or by vacuum-packaging (Campus and others 2008).

Oxidation acceleration in meat after a high-pressure treatment seems to be independent of the oxygen concentration found in the package during the high-pressure treatment, but not during subsequent storage (Cheah and Ledward 1996). These authors showed that oxidation was accelerated in minced pork that was treated in nitrogen gas and then exposed to air in comparison with untreated meat, and the extent of oxidation was the same as for the minced meat treated in air.

According to Tume and others (2010), high-pressure processing accelerates the oxidation of lipid peroxides, effectively reducing their concentration while increasing later breakdown products (detected as TBARS). Since effectively inhibited by EDTA, in-

creases in the rate of lipid oxidation after high-pressure treatment were attributed to the pressure-induced release of iron ions (from myoglobin or ferritin) that catalyze lipid oxidation (Cheah and Ledward 1997; Angsupanich and Ledward 1998). However, Orlien and others (2000) explored the influence of high-pressure treatment on the catalytic activity of metmyoglobin and the effect of free iron in lipid oxidation of raw chicken breast. They concluded that the effect of high pressure (>500 MPa) on lipid oxidation was probably the result of membrane damage in the muscle as this oxidation could not be explained by either a conformational change in metmyoglobin or a release of free iron during the high-pressure treatment. In fact, Carballo and others (1997) found that high-pressure treatment favors the rupture of adipocytes in beef patties. Other minor mechanisms may also contribute to the oxidation chemistry in high-pressure-treated meat products. Serra and others (2007b) demonstrated that high pressure can slightly reduce antioxidant enzyme activity in dry-cured hams. Furthermore, oxidation is not only due to interactions of fat with other meat constituents because a treatment at 800 MPa for 20 min at 19 °C was shown to be catalytic for oxidation in isolated pork fat with an  $a_{w}$  ranging from 0.4 to 0.55 (Cheah and Ledward 1995).

Muscle proteins are also susceptible to oxidative reactions that involve the loss of essential amino acids and decrease protein digestibility, thus affecting the nutritional value of the meat. Moreover, color and texture deterioration of meat has been related to the protein oxidation phenomenon (Xiong 2000). The mechanisms and reaction pathways for the oxidation of lipids and proteins are different but are directly linked as both processes may be affected by similar prooxidant and antioxidant factors. Indeed, it seems that protein oxidation is observed under the same pressure levels as those found when lipid oxidation occurs (>300 MPa). Oxidation of ferrous myoglobin to ferric metmyoglobin was observed in minced beef treated at 400 MPa for 10 min at 10 °C, but was not significant when beef was treated at 200 to 300 MPa (Carlez and others 1995). Myofibrillar proteins in dry-cured ham were oxidized, as shown by the appearance of 2 specific semialdehydes measured by chromatography, after a high-pressure treatment at 600 MPa and 12 °C for 6 min and 1 mo of chilled storage (Fuentes and others 2010). However, in dry-cured ham and loins treated between 200 and 300 MPa for 15 to 30 min at 14 °C, no protein oxidation, estimated by measuring carbonyl, was detected during 90 d of chilled storage, whereas lipid oxidation occurred under the same conditions (Cava and others 2009).

High-pressure treatment of raw and processed meat induces lipid oxidation. Depending on the working pressure and, to a lesser extent, on the treatment duration, lipid oxidation most often occurs during the subsequent storage. Moreover, high pressure can be as damaging as heat treatment in terms of the oxidation level in cold-stored meat products. Generally, high pressure has little effect on lipid oxidation below 300 MPa, but it can have a significant effect at higher pressures.

### High-pressure effects on color and sensory quality of meat and meat products

**Color.** Color is one of the most important attributes of fresh meat that consumers use as a purchasing criterion (Faustman and Cassens 1990). It is well known that high pressure influences raw meat's color. An increase in lightness ( $L^*$ ), for pressures above 200 MPa, is the most often reported modification of raw meat color (Korzeniowski and others 1999; Beltran and others 2004a; Del Olmo and others 2010; Marcos and others 2010). The increase in  $L^*$  results in a whitening effect and has been observed in chicken

meat treated at 400 to 500 MPa and 5 to 10 °C (Beltran and others 2004a; Del Olmo and others 2010), in pork meat treated at 200 to 400 MPa and 20 °C (Korzeniowski and others 1999), and in beef meat treated at 200 to 600 MPa and 10 °C (Carlez and others 1995; Marcos and others 2010). This whitening effect has been related to either (i) protein coagulation with a resulting loss of solubility of sarcoplasmic and/or myofibrillar proteins that affect structure and surface properties (Goutefongea and others 1995); or (ii) globin denaturation and heme group displacement or release (Carlez and others 1995). In beef muscle, a significant effect of high pressure on the  $a^*$  value has also been observed. Jung and others (2003) performed pressure treatments (50 to 600 MPa, 10 °C, 5 min) on raw beef muscle and reported that an increase in pressure up to approximately 350 MPa led to an increase in  $a^*$  values and that these values then decreased at pressures up to 600 MPa. These authors attributed the increase in  $a^*$  values at pressures below 300 MPa to the possible activation of the enzymatic system responsible for metmyoglobin reduction. The decrease in  $a^*$  values at pressures above 350 to 400 MPa was also observed by Marcos and others (2010) and Carlez and others (1995) for beef meat that was high-pressure-treated at 10 to 30 °C. This decrease was linked to the oxidation of ferrous myoglobin to ferric metmyoglobin (Carlez and others 1995; Jung and others 2003; Cava and others 2009) and possibly to further denatured myoglobin ferric species (Wackerbarth and others 2009).

Meat discoloration is significantly, but only slightly, influenced by the treatment duration and can be observed after only 1 min of pressure exposure (Jung and others 2003; Del Olmo and others 2010). The extent of color alteration due to high-pressure treatment in beef patties (Carballo and others 1997) and in pork sausages (Jiménez-Colmenero and others 1997) has been shown to be influenced by fat content. High fat content (20 to 25%) was associated with greater color changes, particularly regarding increased lightness, in comparison with low fat content (9%). Although high-pressure treatments induced visible modifications in raw meat's color, the color difference was greatly reduced after cooking (Gola and others 2000; Mor-Mur and Yuste 2003). Thus, pressure processing of fresh meat cannot be envisaged unless cooking is complete before the final product is presented for sale and consumption (Jung and others 2003; Beltran and others 2004a).

The color of cured products is less affected by pressure than is raw meat's color (Karlowski and others 2002; Rubio and others 2007b), although significant changes have also been reported. For example, a decrease in redness in dry-cured ham was observed above 200 MPa (Andrés and others 2004; Andrés and others 2006; Cava and others 2009). Cava and others (2009) observed that color differences between high-pressure-treated (300 MPa for 30 min at 14 °C) and untreated dry-cured ham and loin disappear during subsequent storage at 4 °C for 90 d. Changes induced by pressure were noticeable after the treatment; however, after 60 and 90 d of storage, differences were no longer found between treated and untreated products. To avoid color changes in cured meat products, cooking before high-pressure treatment has again been recommended (Goutefongea and others 1995). Resistance of the nitrosylmyoglobin pigment to oxidation is probably the reason for the color stability of cured meat products that use nitrates or nitrites (Goutefongea and others 1995; Farkas and others 2002; Pietrzak and others 2007). The effects of drying alone on pressure modifications are not well known. Serra and others (2007a) measured the quality parameters of cured hams that were high-pressure-treated at the beginning or at the end of the resting stage. Pressure induced the appearance of a cooked aspect only in green hams. No

significant modifications were reported in pressure-treated hams at the end of the resting stage, but this different behavior in comparison with green hams was not linked to different  $a_w$  values or moisture contents in the two types of ham. However, according to Comaposada and others (2009), it appears that, in dry-cured meat with moisture contents ranging from 43 to 66%, color changes appear to be smaller in products with reduced water contents. The use of sugars and polyols in the formula of meat products also increases muscle protein stability under pressure (50 to 200 MPa for 20 min at 5 °C) and could limit the development of a cooked appearance under pressure when it is undesirable (Ashie and others 1999). Conversely, smoking does not prevent color changes during pressure treatment (Karlowski and others 2002).

**Aroma and taste.** The development of aroma compounds in high-pressure-treated meat has rarely been investigated. Schindler and others (2010) investigated the effects of high-pressure processing (400 to 600 MPa for 15 min at 5 °C) on the aroma profiles of raw meat (chicken and beef) by measuring the volatile compounds that were released upon opening the package and by sensory evaluation of the odor. The authors reported that, upon opening the bags after 14 d of storage, the sensory trained panel detected an unpleasant off-flavor in the untreated raw beef and chicken that is characteristic of microbial spoilage; however, for the pressure-treated chicken and beef, weak odors typical of the respective meat type emerged during orthonasal flavor perception. It was concluded that pressure treatment of vacuum-packed beef and chicken below 600 MPa does not induce significant changes in the raw aroma profile of either meat type during 14 d of chilled storage in comparison with fresh untreated meat. Rivas-Cañedo and others (2011) evaluated the volatile profile of high-pressure-treated (400 to 600 MPa, 12 °C, 5 to 10 min) cooked pork meat and showed that, during the first 14 d of refrigerated storage, the volatile fractions of pressure-treated samples remained unaltered, whereas those of control samples underwent significant changes. However, beginning on day 14, large differences in the volatile fractions between control samples at day 0 and pressure-treated-samples and between pressure treatments were observed. These differences could be linked to changes in microbiota and were most likely a result of the differences in barosensitivity between bacterial species and even strains.

The development of meat flavor is also linked to the presence of amino acids and peptides that can react during cooking to form Maillard aroma compounds. High pressure (300 to 400 MPa, 20 °C, 10 min) was shown to reduce the content of volatile compounds originating from the Maillard reaction in dry-cured loin (Campus and others 2008).

Finally, high pressure may influence the taste development of meat. High pressure between 100 and 300 MPa for 10 min at 25 °C increases the overall autolytic activity of raw meat and leads to a higher concentration of free amino acids (Ohmori and others 1991). However, with higher pressure treatments at 400 MPa and 500 MPa, the concentration of free amino acids was identical to that of the control sample during one week of chilled storage. In dry-cured loins, Campus and others (2008) showed that high pressure (300 to 400 MPa for 10 min at 20 °C) can stabilize the free amino acid content during storage due to reduction in the activity of aminopeptidases. And, according to Suzuki and others (1994), high-pressure treatments (200 to 400 MPa at ambient temperature) do not influence subsequent changes in the components responsible for the flavor of meat (the amount of amino acids, peptides, and nucleotides) during 7 d of chilled storage. In Japan, it is important that the production of inosinic monophosphate (IMP)

in meat (which is one of the components responsible for “umami” taste) is not delayed by the pressure treatment just after slaughter. The IMP content and the activity of the adenosine triphosphate deaminase, which converts adenosine monophosphate to IMP, was studied by Mori and others (2007) in rabbit muscles treated with high pressure at animal death. They found that the IMP content was instantly increased by a high-pressure treatment of 300 MPa for 5 min at 4 °C and that adenosine triphosphate deaminase activity was maintained at 70% of its initial activity. Lower pressures had almost no effect on IMP production, and the authors concluded that high pressure does not appear to reduce desirable taste characteristics in comparison with conditioned meat. Interestingly, another taste characteristic is also modified by high-pressure treatment. The perception of saltiness detected by a trained panel was increased in high-pressure treated dry-cured ham compared to untreated products (Saccani and others 2004; Fulladosa and others 2009; Clariana and others 2011), possibly due to modified interactions between Na<sup>+</sup> and proteins (Clariana and others 2011).

**Sensory acceptance.** The sensory acceptance of a meat product by consumers can be affected by color modifications, lipid and protein oxidation (Rubio and others 2007b), modification of water retention (Korzeniowski and others 1999) and texture (Uenaka and others 2006; Chattong and Apichartsrangkoon 2009), or development of aroma and taste components (Mori and others 2007; Schindler and others 2010). Furthermore, Sorenson and others (2011) noted that the sensory acceptance of high-pressure-treated meat products depends on consumers’ attitudes toward the purchase and preparation of food (for example, their frequency of ready-made meal consumption). The effect of high pressure on sensory acceptance of meat products varies. Hayman and others (2004) compared consumer acceptance of untreated (7 d of chilled storage) and pressure-treated (600 MPa, 20 °C for 80 s with 98 d of chilled storage) commercial ready-to-eat beef products, Strassburg beef and Cajun beef. They reported no difference in consumer acceptance between untreated products at day 7 of chilled storage and pressure-treated products after 98 d of chilled storage. Rubio and others (2007b) studied the sensory acceptance by consumers of sliced dry-cured beef that was vacuum-packed and stored at 6 °C for 210 d after high-pressure treatment (500 MPa, 5 min) in terms of initial characteristics of the fresh products before packing. Sensory acceptance was not modified immediately after the treatment; however, due to the presence of an anomalous odor and taste, it decreased to the acceptability limit (a score of less than 3 on a 5-point hedonic scale) after 90 d of storage. Thus, the treatment did not improve the shelf-life of this product, even though the product had a lower microbial count than the control. Rivas-Cañedo and others (2011) evaluated the sensory acceptance of the odor of high-pressure-treated (400 to 600 MPa, 12 °C, 5 to 10 min) cooked pork meat and showed that odor acceptability decreased in the same way in control and pressure-treated samples during refrigerated storage.

According to sensory evaluation, raw visceral meat (bovine liver, chicken liver, and gizzards) can be treated at 400 MPa at room temperature for 30 min without losing its tenderness and taste. However, deterioration in these attributes was observed for treatment at 500 MPa (Uenaka and others 2006).

For cooked sausages that were high-pressure-treated ( $\geq 500$  MPa) at 50 to 65 °C, treated samples were generally preferred to conventionally heat-pasteurized products (80 to 85 °C for 40 min) because of their better appearance, taste, and especially texture (Mor-Mur and Yuste 2003; Chattong and Apichartsrangkoon 2009). Interestingly, salt content can be decreased from

2.5% to 1.5% in frankfurter sausages, with an improvement in overall acceptability, by subjecting the meat batters to high pressure at 150 MPa before manufacture and cooking (Crehan and others 2000). And in low-salt beef sausage with 1% NaCl, all of the sensory attributes evaluated with a trained panel were improved by using batter that had been submitted to a high-pressure treatment at 200 MPa for 2 min at 10 °C before cooking (Sikes and others 2009).

Sensory acceptance of high-pressure-treated meat products depends on color, texture, aroma, and taste modifications induced by the process. Problems of sensory acceptance occur with raw products, mainly because of visible color changes. Cooked and cured products are less modified by pressure, and good sensory acceptance is generally reported despite some changes in aroma and taste profiles.

### Chemical safety of high-pressure-treated meat products

The high-pressure treatment process results in a decrease of the microbial load found in meat products and thus increases shelf life. This effect has been demonstrated repeatedly and will be thoroughly reviewed later. In this section, we are interested in the safety of meat products related to toxic compounds, toxins and allergens.

**Toxic amines.** Some amines present in meat products, and particularly in dry-cured or fermented meat products, exhibit toxic effects (Bardócz 1995; Ruiz-Capillas and Jiménez-Colmenero 2004). These amines have been classified as natural polyamines or biogenic amines depending on their synthesis (Bardócz 1995). Natural polyamines are naturally produced by animals and consist of spermidine, spermine, and putrescine. Biogenic amines (histamine, tyramine, putrescine, cadaverine, . . .) only form in the presence of bacterial decarboxylase enzymes, so their concentration in meat products strongly depends on the product microbiota. Lactic acid bacteria (LAB) appear to be the main producers of biogenic amines in fermented products (Halász and others 1994). Toxic amine formation requires the presence of free amino acids, the decarboxylase enzyme, and suitable environmental conditions. Hence, all of the factors bearing on production of free amino acids, the enzymes, and their level of activity affect the type and amount of biogenic amine content (Ruiz-Capillas and Jiménez-Colmenero 2004). These factors may include meat processing. Nevertheless, few studies have investigated biogenic amine content in high-pressure-treated meat products. Pressure treatment (350 MPa, 20 °C, 15 min) caused a weak, but significant, decrease in tyramine, putrescine, and cadaverine levels in dry-cured chorizo during storage, coinciding with a decrease in microbial count, whereas there was a significant increase in spermidine levels that was independent of the bacterial count reduction (Ruiz-Capillas and others 2007b). Inhibition of putrescine and cadaverine accumulation was obtained by preventing *enterobacteria* growth with a high-pressure treatment at 200 MPa for 10 min at 17 °C before sausage fermentation (Latorre-Moratalla and others 2007). In vacuum-packed cooked sliced ham, high-pressure treatment (400 MPa, 30 °C, 10 min), with subsequent storage at 2 °C reduced the initial microbial load and delayed bacterial growth, thus preventing the microbial formation of biogenic amines (Ruiz-Capillas and others 2007a). The same effect of high pressure was observed with pork meat batter treated at 300 MPa and 20 °C for 30 min; in this case, high pressure significantly decreased the contents of spermine, agmatine, and tyramine (Ruiz-Capillas and others 2006). Finally, it has been shown that commercialized high-pressure-treated products generally show lower levels of biogenic amine compounds (associated with lower microbial counts) than



the same products without treatment (Ruiz-Capillas and Jiménez-Colmenero 2004).

**Nitrite-derived substances.** Concerning other possible toxic compounds present in meat, the study of pressure effects on nitrite-derived substances is of particular interest because many high-pressure-treated cured meat products are now commercially available (Garriga and Aymerich 2009). Nitrites can react with amines and amides to form carcinogens. Changes in residual nitrite in high-pressure treated pork products have been studied by Karlowski and others (2002). The authors claim that the physicochemical characteristics of ham, including the residual nitrite level, are not changed by high pressure (300 to 500 MPa, 20 °C, 10 to 30 min) during 8 wk of chilled storage. In pork meat batter, however, Ruiz-Capillas and others (2006) showed a decrease (in comparison with untreated samples) in residual nitrite content when the batter was treated at 300 MPa at 7, 20, or 40 °C for 30 min, and this decrease was enhanced at higher temperatures. The authors also reported that pressure seemed to have no effect on the conversion of nitrite to nitrate, but it significantly decreased the content of protein-bound nitrite compounds.

**Transmissible spongiform encephalopathy agents.** Meat products including bone or brain parts constitute a particular risk for infection by prions and other transmissible spongiform encephalopathy (TSE) agents. Prions are highly resistant and would require autoclaving conditions to be reduced in meat. High pressure has the power to destabilize protein conformation and can dissociate both native and nonnative oligomers. Brown and others (2003) significantly reduced prion infectivity in hot dogs by applying several ultra-high pressure pulses (690 to 1200 MPa) at high temperatures (121 to and temperature (1000 to 1250 MPa, 135 to 142 °C) could also be applied to meat products that contain other TSE agents (Cardone and others 2006). However, according to Heindl and others (2008), TSE infectivity can already be decreased by up to 6 to 8 log units with shorter treatments (5 min) at 800 MPa and 80 °C. The authors stated that the decrease in infectivity with this treatment was faster than prion protein degradation and that these treatments may be less aggressive than heat treatments that are known to denature prion proteins.

**Migration of packaging compounds.** Hugas and others (2002) noted that when using new preservation technologies involving the use of packaging, it is important to study the safety of the material and the possible formation of compounds that influence the aroma and taste of packaged food, as well as the mechanical and physical properties of the material, such as strength and barrier properties. Some studies have shown that the polymer packaging material that is commonly used as a food contact layer is modified by high-pressure treatments. In the study by Dobiás and others (2004) some migration characteristics and the heat sealability of polymer packaging materials were significantly altered by a high-pressure treatment at 600 MPa for 60 min; however, the overall migration limit given by EU legislation was not reached for any of the materials tested. Rivas-Cañedo and others (2009) studied the migration of packaging compounds into high-pressure-treated beef and chicken breast packed in multilayer polymeric bags. A significant migration of compounds from the plastic material was observed, but it was not enhanced by the high-pressure treatment (400 MPa, 10 min, 12 °C). Traces of *n*-hexanal and some hydrocarbons were also found by Schindler and others (2010) while studying volatile compounds in vacuum-packed (in polyethylene bags) high-pressure-treated chicken breast (400 to 600 MPa, 15 min, 5 °C). These authors did not specify whether differences were observed between high-pressure-treated and untreated samples,

but they presumed that the compounds were released from the packaging material.

**Allergens.** The impact of high pressure on allergenicity in high-pressure-treated meat products has rarely been studied (Han and others 2002; Hajós and others 2004). Conformational changes of proteins may alter antigenicity and/or immunological cross-reactivity by changing the binding abilities of their epitopes. Thus, high pressure might either reduce allergenicity or reveal new antigenic sites. Han and others (2002) studied the effect of high-pressure treatment (200 to 600 MPa for 5 min at 5 °C) on the antigenicity of bovine gamma-globulin in beef extract and concluded that high pressure was inefficient in decreasing allergenicity of beef extracts. According to Hajós and others (2004), high pressure (600 MPa for 20 min at ambient temperature) induces conformational changes in pork batter proteins with alteration of some of the epitope structures. The authors observed that, in high-pressure treated samples, some protein groups lose their immunoreactivity, others keep the same level of immunoreactivity, and pressure creates one group of slightly immunoreactive species. Thus, high pressure might also form new protein aggregates with weak immunoreactivity.

Information relating to the effects of high pressure on toxic compounds, toxins and allergens, in meat products is rare. More studies are needed, particularly because the application of high-pressure and high-temperature treatments to different meat products is becoming more commercially important. The few existing studies indicate that high pressure does not enhance the migration of compounds from the packaging material and displays only a small effect on protein allergenicity in meat. Furthermore, high pressure decreases the content of biogenic amines in meat. Thus, high pressure seems to be a safe technology for the treatment of meat products.

### High-Pressure Effects on Microbial Inactivation in Meat and Meat Products

The main purpose of using high pressure to treat meat products is to improve microbial safety. The effects of high pressure on microorganisms are well known and accepted. According to recent studies, cell membrane alterations are probably the main cause of cell death (Patterson 2005; Moussa and others 2009). Ribosome dissociation has also been shown to limit cell viability at high pressure (Abe 2007). Yeasts and molds are more pressure-sensitive than are bacterial vegetative cells (Patterson 2005). However, ascospores are sometimes extremely resistant to high pressure (Chapman and others 2007). Cell barosensitivity is highly dependent on the microorganism species and its growth conditions (Martin and others 2004). Destruction of bacterial spores under pressure is more complex. Different combinations of temperature, time, pressure, and cycling treatments can be used to achieve complete spore inactivation (Farkas and Hoover 2000; Torres and Velazquez 2005). Conversely, relatively moderate pressure levels (200 to 300 MPa) are sufficient to destroy most food parasites (Gamble and others 1998; Molina-Garcia and Sanz 2002; Lindsay and others 2006; Lindsay and others 2008; Porto-Fett and others 2010).

The most important problem encountered in high-pressure pasteurization of food products is that the pressure resistance of microorganisms is reinforced in nutrient-rich media. A significant increase in D-values can be observed for microorganisms inoculated in food in comparison with microorganisms in the buffer (Panagou and others 2007; Tassou and others 2007). Generally, the pressure scale must be higher for food products and validation of treatment parameters using real products is therefore required.

### Influence of the product on microbial inactivation under pressure

Various types of meat products have been high-pressure-treated to extend their shelf-lives. Garriga and Aymerich (2009) distinguish between studies performed on raw meat products and those performed on cooked, cured, and fermented meat products. Table 1 highlights the variability of the high-pressure effect, which depends on both the bacterial species and the type of meat product. However, the results are difficult to compare because processing conditions and methods vary between studies.

The composition of the food matrix has been shown to influence the lethality of a high-pressure treatment despite the fact that the effect of each food constituent on pressure resistance is not well known. First, the microbial reduction is always lower in food than in a buffer system (Smelt 1998; Patterson 2005; Considine and others 2008), and the D-values are increased in ham (Tassou and others 2007) and fish (Panagou and others 2007) in comparison with a PBS buffer. It has also been demonstrated that a low  $a_w$  decreases the efficiency of high-pressure treatments. In products with  $a_w \leq 0.92$  cells are protected against pressure (Garriga and others 2004; Ruiz-Capillas and others 2007b). For example, in the study by

Jofré and others (2009b), dry-cured ham with  $a_w$  of 0.918 showed lower inactivation levels of inoculated microorganisms after high-pressure treatment (600 MPa, 6 min, 31 °C) than did cooked ham and marinated beef loin. However, this undesirable protective effect against pressure seems to be compensated for by the inhibition of the recovery of the cells during storage (Jofré and others 2009b). For example, in cooked ham ( $a_w = 0.98$ ) and fermented sausages ( $a_w = 0.90$ ), the same high-pressure treatment (400 MPa, 10 min, 17 °C) produced completely different results. While the high-pressure treatment produced an immediate 1.8-log CFU/g reduction of *L. monocytogenes* spiked into cooked ham (Aymerich and others 2005), the pathogen was not significantly reduced in fermented sausages (Jofré and others 2009a). However, a progressive decrease in the counts of *L. monocytogenes* was observed during subsequent storage of this latter product, probably due to the progressive death of sub-lethally injured bacteria. Pressure resistance of microorganisms at a low  $a_w$ , like heat resistance in the same conditions, is probably due to the more stable state of macromolecules at low water content (Corry 1975). Furthermore, the  $a_w$  of a food product is also dependent on the concentrations of solutes, such as sugar and salt, the nature of which can

**Table 1—Recent results obtained for microbial inactivation (given as a comparison between untreated control and pressure-treated samples at the same time after treatment) in different meat and meat products treated with high-pressure processing.**

	Meat or meat product	Treatment	Reduction (log CFU/g)	Reference
Raw meat and products with high $a_w$	Raw chicken meat	375 MPa, 18 °C, 15 min	Inoculated <i>Listeria monocytogenes</i> : 2 to 5 immediately after processing, depending on the strain	Simpson and Gilmour (1997)
	Poultry sausages	500 MPa, 50 °C, 10 min 500 MPa, 60 °C, 10 min	Aerobic mesophiles: 3.28 the day after processing 5.18 the day after processing	Yuste and others (2000b)
	Mechanically recovered poultry meat	450 MPa, 20 °C, 15 min	Aerobic mesophiles: 3.7 the day after processing	Yuste and others (2001)
	Frankfurt sausages	500 MPa, 65 °C, 15 min	Aerobic mesophiles: 6.14 after 3 wk of chilled storage	Yuste and others (2000a)
	Raw smoked pork loin	500 MPa, 30 min	Aerobic mesophiles: 1.36 just after processing 0.5 after 8 wk of chilled storage	Karlowski and others (2002)
	Raw beef	560 MPa, 10 °C, 4 min	Aerobic mesophiles: 2.5 the day after processing	Jung and others (2003)
	Marinated beef loin	600 MPa, 31 °C, 6 min	Aerobic mesophiles: 6.51 (of 6.51 initially present) immediately after processing	Garriga and others (2004)
	Marinated beef loin	600 MPa, 31 °C, 6 min	Inoculated LAB: about 4 of 5 initially present, 2 d after processing	Jofré and others (2009b)
Dry cured products	Dry cured ham	600 MPa, 31 °C, 6 min	Aerobic mesophiles: 2.7 immediately after processing	Garriga and others (2004)
	Dry cured ham	600 MPa, 31 °C, 6 min	Inoculated LAB: 1.6 (of 4 initially present), 2 d after processing	Jofré and others (2009b)
	Dry fermented pork sausage	400 MPa, 17 °C, 10 min	Inoculated <i>Listeria monocytogenes</i> : 0.6 (of 6 initially present) the day after processing	Jofré and others (2009a)
	Dry cured chorizo sausages	350 MPa, 20 °C, 15 min	Aerobic mesophiles: <1 immediately after processing	Ruiz-Capillas and others (2007b)
	Dry cured beef "Cecina de Leon"	500 MPa, 18 °C, 5 min	Aerobic Mesophiles: 1.66 after processing 2.55 after 60 d of storage	Rubio and others (2007b)
Cooked meat and products (low acid, high $a_w$ )	Sliced cooked ham	400 MPa, 7 °C, 20 min	Aerobic mesophiles: 2.34 immediately after processing	Lopez-Caballero and others (1999)
	Sliced cooked ham	300 MPa, 20 °C, 15 min	Aerobic mesophiles: 0.3 immediately after processing	López-Caballero and others (2002a)
	Sliced cooked ham	400 MPa, 17 °C, 10 min	Inoculated <i>Listeria monocytogenes</i> : 1.8 immediately after processing	Aymerich and others (2005)
	Cooked ham	600 MPa, 31 °C, 6 min	Aerobic mesophiles: >2.45 immediately after processing	Garriga and others (2004)
	Cooked ham	600 MPa, 20 °C, 10 min	Aerobic mesophiles: 1.5	Karlowski and others (2002)
	Blood sausages	600 MPa, 15 °C, 10 min	Aerobic mesophiles: 2.62 the day after processing	Diez and others (2008)
	Frankfurters	400 MPa, 30 °C, 10 min	Total viable count: 2.16 immediately after processing	Ruiz-Capillas and others (2007c)
Fried minced pork meat	400 MPa, 20 °C, 60 min	Inoculated <i>Bacillus stearothermophilus</i> : 2 immediately after processing	Moerman and others (2001)	

significantly affect cell survival after a pressure treatment (as discussed in the reviews of Considine and others 2008 and Smelt 1998). The barotolerance observed at elevated levels of osmolarity could be due to the microbial uptake of compatible solutes (such as betaine or carnitine) from the external environment (Smiddy and others 2004). These solutes play the role of stabilizers for enzyme functions or of osmotic balancers (Hill and others 2002). However, this has only been shown in studies carried out in model media.

Park and others (2001) compared the inactivation of *Lactobacillus viridescens* in Man Rogosa Sharp (MRS) and in protein-fortified MRS broth and reported that the addition of proteins decreased the inactivation after a high-pressure treatment at 400 MPa for 5 min at 20 °C. However, these authors did not provide the  $a_w$  values of the media tested. The influence of nutrient composition on microbial inactivation was evaluated by Moerman and others (2001) who compared the high-pressure-induced reduction of different microorganisms in fried chicken and in mashed potatoes (two products with similar pH values of 5.9 and  $a_w$  values of 0.98) using an experimental design defined over 0 to 400 MPa, 20 to 80 °C and 1 to 60 min. The effect of the medium was shown to be negligible and, thus, this study did not reveal any protective action of the major nutrient fractions (carbohydrates, fat, and proteins). Nevertheless, the microbial protection of fat has been mentioned several times. Rubio and others (2007a, b) observed that high pressure (500 MPa for 5 min at 18 °C) did not produce an inhibitory effect on the mesophilic count in dry sausages, whereas the same treatment efficiently delayed the growth of spoilage microorganisms in dry-cured beef. These authors hypothesized that the different behaviors of microorganisms in dry sausages and dry-cured beef might be caused by the protective effect of fat in dry sausages. However, no significant difference in the reduction of the total aerobic count was observed between low-fat (90 g/kg) and high-fat sausages (247 g/kg) that were treated for 20 min at 300 MPa (Jiménez-Colmenero and others 1997). According to Escriu and Mor-Mur (2009), this effect is dependent on both the type of fat and the type of microorganism tested. These authors showed that immediately after a high-pressure treatment (400 MPa, 20 °C, 2 min), the *Listeria innocua* count was more reduced in chicken meat mixed with olive oil than in the same meat mixed with tallow. In addition, depending on the type and percentage of fat content, *Listeria innocua* and *Salmonella Typhimurium* did not recover in the same way after 60 d of cold storage; however, it was not possible to show any clear relationship with either fat content or fat quality. Rubio and others (2007a) also failed to establish a clear relationship between the fatty acid composition of a meat product and the effectiveness of high-pressure treatment. These authors evaluated the microbiological quality of 3 types of sausages with different compositions of fat (control, high oleic, and high linolenic) after treatment at 500 MPa for 5 min at 18 °C. Thus, it appears that the effect of fat is not simple and may depend on its composition, its location in the product, and its interactions with the other components of the matrix. For example, olive oil may contain antimicrobial phenolic compounds (Medina and others 2007) that could explain the higher cell reduction observed in meat supplemented with olive oil in comparison with meat supplemented with tallow (Escriu and Mor-Mur 2009).

Cell recovery during subsequent storage is another important item to consider. Many authors have reported low recovery of microorganisms in high-pressure-treated products during subsequent storage, and, in most cases, bacterial growth is delayed by a high-pressure treatment at a sufficient pressure level ( $\geq 400$  MPa) (Yuste and others 2000a; Garriga and others 2004; Jofré and others

2009b). For example, Patterson and others (2010) showed that the total viable count in cooked poultry meat treated at 600 MPa at 18 °C for 10 min could be stabilized to 3 log CFU/g during 35 d of cold storage. Sometimes the microbial count is not reduced immediately after the pressure treatment but shows a significant decrease during cold storage, as for *enterobacteria* in blood sausages treated at 300 to 600 MPa for 10 min at 15 °C (Diez and others 2008) or for *S. aureus* spiked in dry-cured ham treated at 600 MPa for 6 min at 31 °C (Jofré and others, 2009b). However, for cooked products, problems of fast microbial recovery during subsequent storage have sometimes been described (Garriga and others 2002, 2004). Recovery of *Escherichia coli* and LAB reached the level of the control after less than 20 d of chilled storage in cooked ham homogenized with water and treated at 400 MPa for 10 min at 17 °C (Garriga and others 2002). In sliced cooked ham high-pressure-treated at 600 MPa for 6 min at 31 °C, total aerobic count increased during chilled storage, mainly due to LAB growth, but no recovery was observed in dry-cured ham or in marinated beef loin treated in the same conditions (Garriga and others 2004). This may be due to the negative effect of high pressure on the water-holding capacity of cooked products that thus produce rich exudates (Pietrzak and others 2007) and to the fact that cooked products do not display hurdles against microbiological growth during cold storage (Garriga and Aymerich 2009). Besides permitting a fast recovery, rich exudates could also display a protective effect against inactivation as a higher survival of *Listeria monocytogenes* was observed in cooked chicken and beef mince in comparison with raw meat (Simpson and Gilmour 1997).

Microbial reduction and recovery during storage in high-pressure-treated food products depend on the type of product tested. In products with low  $a_w$  ( $\leq 0.9$ ) cells are protected against pressure, but recovery is inhibited during storage. In cooked products, fast recovery during subsequent storage can be observed. There is a significant impact of food composition on microbial reduction and the effect of protein and fat is complex. Finally, differences in lethality and recovery rates may be due to how the food matrix tolerates pressure treatments and to the ways in which the interactions of all components affect this matrix and are not restricted to the pressure effects on a single component.

### Influence of the microorganism species on the inactivation under pressure

*Enterobacteria* and LAB are the main components of the deterioration flora in meat. Low-temperature alterations are provoked by psychrotrophs (mainly *Pseudomonas*). Concerning pathogenic microorganisms, *Salmonella*, *Listeria monocytogenes*, and some specific strains of *Escherichia coli* are the greatest threats. Additionally, in industrialized countries, *Campylobacter* spp. (especially *jejuni*) are a major cause of enteritis and are found mainly in poultry and to a lesser extent in pork (Belloc and others 2004). The risk of the development of the dreaded toxin-producing anaerobic *Clostridium botulinum* continues to exist with long-term storage at high temperature (25 to 40 °C) of some meat products.

Recent results of bacterial reduction in meat products are presented in Table 2 for alteration flora and in Table 3 for pathogenic flora. A high-pressure treatment (400 to 600 MPa, 7 to 18 °C, 5 to 10 min) is generally effective for reducing the number of *enterobacteria* below the level of detection in high  $a_w$  products, such as cooked ham, marinated beef loin, or blood sausages (López-Caballero and others 1999; Garriga and others 2004; Diez and others 2008). *Escherichia coli* is a potentially pathogenic enterobacterium, and the effects of high pressure on some relevant species

Table 2—Recent results obtained for inactivation of alteration flora in meat and meat products treated by high-pressure process (HP = high-pressure-treated sample).

Family	Genus/Species	Gram	Treatment	Meat or meat product	Microbial load (log CFU/g)	Reference
<i>Enterobacteria</i>		—	400 MPa, 5 °C, 5 min	Cooked ham	After 35 d of chilled storage Control: 6.36; HP: <1	Lopez-Caballero and others (1999)
			600 MPa, 16 °C, 6 min	Cooked ham	After 90 d of chilled storage control: 3.71; HP: <1	Garriga and others (2004)
				Marinated beef loin	After 90 d of chilled storage control: 5.16; HP: <1	
		300 MPa, 15 °C, 10 min	Blood sausages	After 28 d of chilled storage: control: 1.92; HP:<1	Diez and others (2008)	
	<i>Pseudomonas</i> sp.	—	450 MPa, 20 °C, 20 min	Fresh minced meat	Control: 5; HP: <1 immediately after high pressure processing, but total recovery after 15 d of chilled storage	Carlez and others (1994)
	<i>P. fluorescens</i> isolated from pork meat		400 MPa, 20 °C, 10 min	Culture broth	Control: 7.8; HP: <1 immediately after high pressure processing, but total recovery after 16 d of storage at 20 °C	Lopez-Caballero and others (2002b)
	<i>Pseudomonas</i> sp.		300 MPa, 15 °C, 10 min	Blood sausages	After 21 d of chilled storage, control: 3.30; HP: <2	Diez and others (2008)
Lactic acid bacteria	<i>Lactobacillus sakei</i> and <i>Leuconostoc carnosum</i>	+	400 MPa, 5 °C, 20 min	Cooked ham	After 7 d of chilled storage, control: 3.78; HP: <1 after 35 d of chilled storage, control : 7.28; HP: 2.66	Lopez-Caballero and others (1999)
			400 MPa, 17 °C, 10 min	Model meat system	Immediately after high pressure processing, control: 8.5; HP: <2 after 6 d of chilled storage, recovery to 6 for HP	Garriga and others (2002)
			600 MPa, 16 °C, 6 min	Cooked ham	After 30 d of chilled storage, control: 7.84; HP: <2 after 120 d of chilled storage, control : 8.71; HP: 2.65	Garriga and others (2004)
				Beef loin	After 120 d of chilled storage, control: 8.68; HP: <2	
			500 MPa, 18 °C, 5 min	Cecina de Leon (smoked and dried beef meat) cuts	After 15 d of chilled storage, control : 3.83; HP: <1	Rubio and others (2007b)
			600 MPa, 15 °C, 10 min	Blood sausages	Immediately after high pressure processing, control: 5.46; HP: 5.41 after 28 d of chilled storage, control : 8.67; HP : 8.50	Diez and others (2008)
	300 MPa, 27 °C, 10 min	Sliced ham	After 20 d of chilled storage, control: 6.5; HP: 2 after 40 d of chilled storage, control : 6.5; HP: 6	Slongo and others (2009)		

have been studied. *E. coli* has also been shown to have a high sensitivity to pressure. In cooked ham, dry-cured ham, and marinated beef loin inoculated at 3.5 log CFU/g, and in marinated beef with an endogenous load of 1.18 log CFU/g, *E. coli* was reduced below the level of detection during 120 d of chilled storage by a high-pressure treatment at 600 MPa for 6 min at 31 °C (Garriga and others 2004; Jofré and others 2009b). In the study of Garriga and others (2002), a mixture of 2 strains of *E. coli* displayed a 4.5-log CFU decline 24 h after a high-pressure treatment (400 MPa, 10 min, 17 °C). A high-pressure treatment of 400 MPa at 12 °C for 20 min was sufficient to give a reduction of 2.45 log CFU/g (of 7 log initially present) of a pressure-resistant strain of the serotype O157:H7 in ground beef (Morales and others 2008). Porto-Fett and others (2010) showed a total reduction of the initial 5 log CFU/g of *E. coli* O157:H7 inoculated into dry-fermented salami after a high-pressure treatment at 483 MPa at 19 °C for 5 min. A total reduction was also reported by Gola and others (2000) in raw minced meat that was inoculated with a mixture of 8 strains of *E. coli* O157:H7 and treated at 700 MPa at ambient temperature for 5 min.

Psychrotrophic microorganisms are also pressure-sensitive and are more susceptible to pressure than are mesophiles. Yuste and others (2001) showed a reduction of 4.74 log CFU/g

for psychrotrophs and of 3.7 log CFU/g for mesophiles in mechanically recovered poultry meat treated at 450 MPa for 15 min at 20 °C. Garriga and others (2004) reported a reduction in the psychrotrophic bacteria in high-pressure treated sliced dry-cured ham and sliced cooked ham (600 MPa, 16 °C, 6 min) to levels below the level of detection during 60 d of subsequent storage, whereas mesophilic bacteria were less reduced by the treatment and recovered during storage. One possible explanation is that when most psychrotrophs are subjected to high pressure, they lose their ability to grow at low temperatures, preventing their recovery during subsequent chilled storage. Psychrotrophs in meat are mainly composed of bacteria from the genus *Pseudomonas* (Jay and others 2003; Ercolini and others 2009). Processing meat at pressures between 300 and 450 MPa appears to be sufficient to completely inactivate indigenous *Pseudomonas* (Carlez and others 1994; López-Caballero and others 2002a). However, after a lag period, cells from *Pseudomonas* spp. can be detected again and resume growth, as was observed by Carlez and others (1994), in fresh minced meat. The same phenomenon was observed by López-Caballero and others (2002b) for *Pseudomonas fluorescens* isolated from pork meat and inoculated at 8 log CFU/g in culture broth. Even with a total reduction just after the pressure treatment (400 MPa, 20 °C,



**Table 3**—Recent results obtained for inactivation of pathogenic flora (given as the reduction immediately after high-pressure treatment) in meat and meat products.

Genus/Species	Gram	Treatment	Meat or meat product	Reduction (log CFU/g)	Reference
<i>Listeria monocytogenes</i>	+	375 MPa, 18 °C, 20 min	Raw chicken mince Cooked chicken mince Model meat system	4 of 8.7 inoculated 1.5 of 8.7 inoculated	Simpson and Gilmour (1997)
		400 MPa, 17 °C, 10 min		6.5 of 8 inoculated, total recovery after 20 d of chilled storage	Garriga and others (2002)
		400 MPa, 17 °C, 10 min	Sliced cooked ham	4 of 4 inoculated and recovery to about 8 log CFU/g after 40 d of chilled storage	Aymerich and others (2005), Marcos and others (2008a)
		500 MPa, 20 °C, 1 min	Turkey breast meat	0.9 of 7 inoculated.	Chen (2007)
		500 MPa, 25 °C, 10 min	Sliced cooked ham	5 of 5 inoculated but total recovery after 70 d of chilled storage	Koseki and others (2007)
		600 MPa, 10 °C, 5 min	Cooked ham	3.5 of 4 inoculated	Jofré and others (2008b)
		600 MPa, 18 °C, 5 min	Salami	1.6 to 6 of 7 inoculated, depending on fermentation and drying conditions. No or slight recovery under chilled storage	Porto Fett (2010)
<i>Listeria innocua</i>		400 MPa, 20 °C, 2 min	Chicken breast	1.5 to 3 depending on the composition	Escriu and Mor-Mur (2009)
<i>Staphylococcus aureus</i>		600 MPa, 31 °C, 6 min	Marinated beef loin Dry cured ham Cooked ham,	2.5 of 3.5 inoculated 0.5 of 3.5 inoculated 1.1 of 3.5 inoculated	Jofré and others (2009b)
		400 MPa, 17 °C, 10 min	Model meat system	No significant reduction of 8 inoculated	Garriga and others (2002)
		400 MPa, 20 °C, 30 min	Pork Marengo	no significant reduction of 4.6 inoculated	Moerman 2005
<i>Salmonella Typhimurium</i>	–	400 MPa, 20 °C, 2 min	Minced chicken	3.26 to 4.35 (depending on the composition), total recovery after 25 d of chilled storage	Escriu and Mor-Mur (2009)
<i>Salmonella enterica</i>		400 MPa, 17 °C, 10 min	Meat model	6 of 8 inoculated. No recovery during 60 d of chilled storage	Garriga and others (2002)
		400 MPa, 17 °C, 10 min	Fermented sausages	2 of 2.7 inoculated. Inactivation to <1 log CFU/g after 20 d of chilled storage	Jofré and others (2009a)
<i>Salmonella enteritidis</i>		500 MPa, 50 °C, 10 min	Poultry sausages	7.16 of 8 inoculated.	Yuste and others (2000b)
<i>Escherichia coli</i> O157 H7		700 MPa, 20 °C, 5 min	Raw minced meat	Total inactivation	Gola and others (2000)
<i>Escherichia coli</i>		400 MPa, 17 °C, 10 min	Model meat system	4.5 of 8 inoculated. Recovery after 10 d of chilled storage	Garriga and others (2002)
Endogenous <i>Escherichia coli</i>		600 MPa, 16 °C, 6 min	Marinated beef loin	Total inactivation of 1.18 initially present during 120 d of chilled storage	Garriga and others (2004)
<i>Escherichia coli</i> O157 H7		400 MPa, 12 °C, 20 min	Ground beef patties	2.45 of 7 inoculated.	Morales and others (2008)
<i>Escherichia coli</i>		600 MPa, 31 °C, 6 min	Cooked ham, dry cured ham and marinated beef loin	Total inactivation of 4 inoculated. Slight recovery only for cooked ham	Jofré and others (2009b)
<i>Escherichia coli</i> O157 H7		483 MPa, 19 °C, 5 min	Dry fermented salami	5 of 5 inoculated	Porto Fett (2010)
<i>Campylobacter jejuni</i>		200 MPa, 25 °C, 10 min	Chicken meat	2 of 8 inoculated	Martinez-Rodriguez and Mackey (2005)
		450 MPa, 15 °C, 1 min	Chicken slurry	7 of 7 inoculated	Lori and others (2007)
		600 MPa, 31 °C, 6 min	Cooked ham, dry cured ham and marinated beef loin	3.5 of 3.5 inoculated	Jofré and others (2009b)

10 min), after 8 d of incubation, the cell counts were similar for high-pressure-treated and untreated samples. In the study by Diez and others (2008) on blood sausages treated between 300 and 600 MPa at 15 °C, *Pseudomonas* was the most pressure-sensitive microbial group (along with *Enterobacteria*), and the bacterial count remained below the level of detection during 28 d of cold storage.

Conversely, the baroresistance of LAB has been widely reported, although the resistance and ability of LAB to recover from a high-pressure treatment can be positive or negative depending on whether the strain is used for its technological properties or is a spoilage strain. Different behaviors of LAB after high-pressure treatment have been observed depending on the strain and the food matrix. When 2 inoculated LAB strains were treated at 400 MPa

at 17 °C for 10 min in a meat model, immediate reductions of 8.5 log were observed. However, after 20 d at 4 °C, both strains reached levels of at least 6 log CFU/g (Garriga and others 2002). Garriga and others (2004) observed the ability of endogenous LAB present in cooked ham to recover during storage of the product at 4 °C after a 600-MPa treatment. In contrast, the authors found no recovery of LAB in high-pressure-treated dry-cured ham and in beef loin during 120 d of storage at 4 °C. In dry-cured beef, a reduction of 3 log CFU was attained one day after a high-pressure treatment at 500 MPa for 5 min at 18 °C (Rubio and others 2007b), and the bacterial growth was delayed during the subsequent chilled storage. In blood sausages, LAB counts were only slightly reduced by a high-pressure treatment at 600 MPa for

10 min at 15 °C (Diez and others 2008). In sliced cooked ham, a significant reduction in the LAB population and a marked delay in their recovery can be obtained with a high-pressure treatment at 400 MPa for 20 min at 7 °C (López-Caballero and others 1999) or for 5 min at 27 °C (Slongo and others 2009). This treatment delays the use-by date from 19 to 85 d despite a recovery of LAB (Slongo and others 2009). All of these data confirm the fact (as mentioned by various authors, such as Patterson 2005 and Escriu and Mor-Mur 2009) that Gram+ bacteria (LAB) are more resistant than gram-bacteria (*Enterobacteria*, *Pseudomonas*); this is probably a result of a more robust cell envelope in Gram+ bacteria that contains a high percentage of peptidoglycan and teichoic acids, as suggested by Escriu and Mor-Mur (2009).

The higher pressure resistance of Gram+ bacteria is confirmed by the comparison of the resistance of the 2 main pathogens of meat, namely *Listeria* (Gram+) and *Salmonella* (Gram-) (Garriga and others 2002; Escriu and Mor-Mur 2009). The contamination of meat products with *Listeria monocytogenes* is a major public health problem. Thus, numerous studies have been devoted to the effect of high-pressure treatment on this pathogen (Simpson and Gilmour 1997; Garriga and others 2002; Hayman and others 2004; Aymerich and others 2005; Chen 2007; Koseki and others 2007; Marcos and others 2008a, b; Porto-Fett and others 2010). Generally, a treatment at 400 MPa is necessary to significantly decrease the *Listeria* load (Simpson and Gilmour 1997; Chen 2007). According to Porto-Fett and others (2010), a reduction of 1.6 to  $\geq 5$  log CFU/g can be achieved in salami by high-pressure treatment at 600 MPa and 18 °C for 1 to 7 min or at 483 MPa and 18 °C for 5 to 12 min, depending on the  $a_w$  of the product and on the treatment strength. Garriga and others (2004) also emphasized the importance of the type of product processed for reducing the safety risks associated with *Listeria monocytogenes*. According to these authors, a pressure of 600 MPa for 6 min at 31 °C is sufficient for sliced marinated beef loin and for dry-cured ham. In chicken batters, containing a different type of fat, a reduction of 1.5 to 3 log CFU/g of *Listeria innocua* was achieved after a high-pressure treatment at 400 MPa and 20 °C for 2 min (Escriu and Mor-Mur 2009). However, regardless of the immediate reductions in the cell count, the main risk associated with *Listeria* is its potential recovery during cold storage. According to Marcos and others (2008a), pressure treatment of cooked ham at 400 MPa and 17 °C for 10 min and cold storage cannot prevent *Listeria* recovery; 600 MPa and 10 °C for 5 min followed by cold storage below 6 °C is necessary to ensure cooked ham's safety for up to 3 mo (Jofré and others 2008b).

The same treatments that can remove *Listeria* risk are also generally effective at inactivating *Salmonella* spp., which show less potential for recovery during subsequent storage (Garriga and others 2002; Jofré and others 2009b). For example, a treatment at 600 MPa for 6 min at 31 °C permitted researchers to reduce a cocktail of inoculated salmonella strains from 3.5 log CFU/g to <10 CFU/g in cooked ham, dry-cured ham, and marinated beef loin (Jofré and others 2009b). A treatment at 500 MPa and 50 °C for 10 min can lead to a reduction of more than 7 log CFU/g in inoculated poultry sausages (Yuste and others 2000b).

Among food-borne pathogens, *Staphylococcus aureus* appeared to be the most resistant to high pressure in comparison with *L. monocytogenes*, *Salmonella*, *Yersinia enterocolitica*, and *Campylobacter jejuni* (Jofré and others 2009b). After the application of a 600-MPa treatment (for 6 min at 31 °C), the reduction in *S. aureus* counts in meat products spiked at 3.5 log CFU/g was 2.7 log units in beef loin and 1.1 log units in cooked ham. In dry-cured ham, the pathogen

only decreased by 0.5 log units after the high-pressure treatment. Treatment at 700 MPa for 5 min was necessary to achieve complete inactivation of the pathogen in a buffer initially inoculated at 4.5 CFU/mL, although no investigation of the possible recovery of sub-lethally injured cells was carried out (Yuste and others 2004). In a meat model, 2 different strains of *Staphylococcus aureus* were resistant to a treatment of 400 MPa at 17 °C for 10 min (Garriga and others 2002), and in pork Marengo, *S. aureus* was affected little by a pressure treatment at 400 MPa and 20 °C for 30 min (Moerman 2005).

Investigations have also been carried out with the Gram- *Campylobacter* and more frequently with *C. jejuni*. Martínez-Rodríguez and Mackey (2005) treated (200 to 400 MPa, 25 °C, 10 min) different *Campylobacter jejuni* strains inoculated into chicken meat. This microorganism has a relatively high sensitivity to pressure depending on the strain, as a 200-MPa treatment could sometimes reduce the microbial count by 2 log CFU/g. The low resistance to pressure of *C. jejuni* was confirmed by Lori and others (2007) who showed that treatment at 450 MPa and 15 °C for 30 seconds was sufficient to inactivate more than 6 log CFU/g in chicken slurry. According to Jofré and others (2009b), *Campylobacter jejuni* was the most inactivated microorganism among *Salmonella*, *Listeria*, *Staphylococcus*, *Escherichia coli*, and LAB that were spiked into different types of meat products. As of yet, no study has addressed *Campylobacter* recovery after a high-pressure inactivation.

Whatever the bacterial species, it must be highlighted that the obtained inactivation is highly dependent on the strain; sometimes a difference of 3 log CFU can be observed in the inactivation of two strains of the same bacterial species for the same treatment (Simpson and Gilmour 1997). This finding suggests the importance of using a cocktail of strains as target bacteria in further studies of meat and meat products (Garriga and others 2002; Jofré and others 2009b). In addition, endogenous microbiota are believed to be more resistant than inoculated collection strains (Carlez and others 1994; Yuste and others 2001).

Increasing numbers of studies now address the development and the composition of product microbiota during storage after high-pressure treatment (Yuste and others 2000a; Garriga and others 2002; Tuboly and others 2003; Patterson and others 2010). Patterson and others (2010) showed that a high-pressure treatment could result in the selection of barotolerant species in meat products, which can result in a beneficial effect on the quality of the product during storage. The authors found that in cooked poultry meat treated at 500 to 600 MPa for 1 to 10 min at 18 °C, only bacteria identified as the LAB *Weissella viridescens* grew during the cold storage. Despite a high *Weissella* count, no signs of spoilage were observed on the pressure-treated samples during 35 d of chilled storage, probably due to antimicrobial activity of the bacteria against a wide range of microorganisms.

Microbial reduction following a high-pressure treatment is highly dependent on the bacteria species. Gram- bacteria (*Enterobacteria*, bacteria of the genus *Pseudomonas* and *Campylobacter*) are generally more pressure-sensitive than Gram+ bacteria (LAB, *Listeria monocytogenes*, *Staphylococcus aureus*). Thus, high-pressure treatment modifies the development and the composition of the microbiota in meat products during their subsequent storage.

The same factors that influence microbial inactivation in high-pressure-treated meat products may also alter the resulting quality of the products. Thus, an essential challenge for the industry is to find strategies to optimize microbial inactivation while providing

meat products with desirable physicochemical qualities and sensory attributes.

## Strategies used to Improve Microbial Inactivation during the High-Pressure Process

### Strategies used to improve high-pressure pasteurization

An increase in the treatment temperature is the most frequently used strategy used to improve the lethality of high-pressure treatments. Elevated temperatures (45 to 60 °C) allow for lower pressures and shorter processing times for pathogen inactivation compared to application at room temperature (Simpson and Gilmour 1997; Yuste and others 2000b; Moerman 2005; Tassou and others 2008). For example, a 5-log reduction of the pressure-resistant *Staphylococcus aureus* can be achieved in chicken treated at 500 MPa at 50 °C for 15 min, whereas a pressure level of approximately 700 MPa would be necessary to obtain the same reduction at 20 °C (Patterson and Kilpatrick 1998). According to Yuste and others (2000a), high-pressure processing at 500 MPa for 5 min at 65 °C could be an effective preservation method to replace heat pasteurization (80 °C, 40 min) in cooked sausages, achieving the same preserving effect in terms of microbial inactivation and growth inhibition during chilled storage. However, heat-transfer limitations in particulate food products can still prevent their successful pasteurization by high-pressure treatments at elevated temperatures (Moerman 2005).

The use of temperatures below 15 °C during high-pressure treatment could also be an effective strategy to improve treatment efficiency. A pork-isolated strain of *P. fluorescens* was reduced by 5 log when treated at 200 MPa and 5 °C for 10 min, and the bactericidal effect decreased as the temperature increased to 35 °C (López-Caballero and others 2002a). In fact, Chen (2007), exploring the effect of pressure at temperatures ranging from 1 to 55 °C, found that *L. monocytogenes* was most resistant to pressure (500 MPa, 1 min) at temperatures between 10 and 30 °C. As temperatures decreased below 10 °C or increased above 30 °C, *L. monocytogenes*' susceptibility to pressure increased. This enhanced inactivation effect was more pronounced when meat samples were treated at higher temperatures. In the study by Yuste and others (2002), a high-pressure treatment at subzero temperatures (450 MPa, -20 °C, 15 min) was not effective at decreasing the microbial count in poultry meat, probably because samples were treated while frozen and thus presented a lack of plasticity. Combining subzero temperatures and high pressure in the liquid state in distilled water has been shown to improve microbial inactivation. In fact, Moussa and others (2006) show that the pressure required to achieve a total inactivation of 8 log CFU/mL of *E. coli* at -20 °C was 250 MPa, whereas it was 350 MPa at 25 °C.

Several authors have studied the potential of using oscillatory high-pressure processing of meat to improve process lethality. Yuste and others (2001) subjected poultry meat to alternating moderate pressure (60 MPa) and high pressure (450 MPa) but did not obtain any increase in efficiency in comparison with a continuous treatment at 450 MPa. According to Morales and others (2008), a multiple-cycle treatment of four 1-min cycles at 400 MPa and 12 °C is as effective at reducing *E. coli* 0157:H7 in ground beef as is a continuous treatment of 20 min at the same pressure. On the contrary, the use of multiple-cycle treatments instead of single-cycle treatment for the same duration at 400 MPa and 12 °C was shown to be advantageous for the inactivation of *Salmonella enteritidis* inoculated onto chicken breast fillets (Morales and others 2009). Because oscillations of pressure can be damaging for high-pressure equipment, an increase in efficiency should be demon-

strated to justify this option. Furthermore, multiple-cycles treatment does not prevent modifications of color and texture under pressure in raw meat. Color changes occur even during very short treatments of 1 min at 400 MPa, and hardness significantly increases in pressure-treated raw fillets under either multiple cycles or continuous pressure (Del Olmo and others 2010).

The synergism between bacteriostatic additives and high pressure has been reported (Krockel and Muller 2002; Aymerich and others 2005; Marcos and others 2008b; Ogihara and others 2009). Krockel and Muller (2002) observed that recovery and growth of microorganisms were much more restricted in Lyoner sausage than in Gelbwurst (a 'diet bologna' without nitrite) in subsequent storage after a high-pressure treatment at 400 MPa and 10 °C for 10 min. The authors attributed this effect to the presence of nitrites in Lyoner sausage. Ogihara and others (2009) studied the synergistic effects of high pressure and 27 different food additives in *Salmonella enteritidis* suspensions and found significant synergistic effects of citric acid, adipic acid, glycerin mono-caprylic acid ester (C8), glycerin mono-capric acid ester (C10), tannic acid, nisin, wasabi extract, e-polylysine, and protamine sulfate. Indeed, organic acids can be used in meat products to delay the growth of bacteria during storage without negative effects on the sensory quality of the products (Diez and others 2008). In blood sausages, the addition of a 3% mixture of potassium and sodium lactates delays the growth of LAB so that after 21 d of chilled storage the LAB count was lower than 7 log CFU/g compared to almost 8 log CFU/g in the control (Diez and others 2008). In sliced cooked ham prepared with 1.8% potassium lactate, the growth of *Listeria monocytogenes* during chilled storage at 6 °C was delayed, and the combination of this treatment with a high-pressure treatment at 400 MPa for 10 min at 17 °C totally inhibited the recovery of *Listeria monocytogenes* during chilled storage at 6 °C (Aymerich and others 2005). Lactate (1.4%) was also efficient at preventing *Listeria monocytogenes* growth after a cold-chain break in high-pressure treated cooked ham (400 MPa for 10 min at 17 °C) (Marcos and others 2008b). In fact, high pressure alone decreased the initial count of *Listeria monocytogenes* from 5 to 2 log CFU/g, but it failed to prevent the bacteria from recovering after a cold-chain break arising during the storage.

Among antimicrobials, bacteriocins have also been used in combination with high pressure to increase microbial inactivation (Yuste and others 1998; Garriga and others 2002; Yuste and others 2002; Marcos and others 2008b). Bacteriocins can be either incorporated into the product formulation or applied directly to meat before the pressure treatment. The use of active packaging containing antimicrobials is an additional solution that has been shown to be effective (Jofré and others 2008a; Marcos and others 2008a). Bacteriocins are antibacterial peptides produced by bacteria that kill or inhibit the growth of other bacteria. Nisin is currently the only bacteriocin to be widely used as a food preservative. Its effectiveness in reducing the *Listeria* risk is low, and the use of potassium lactate has been shown to be more effective at inhibiting cell recovery during chilled storage (Aymerich and others 2005). The mechanism proposed to explain the synergism between high pressure and bacteriocins is that cells surviving the pressure treatment are sub-lethally injured and are then more sensitive to bacteriocins (Masschalck and others 2001). Garriga and others (2002) investigated the behavior of several foodborne bacteria inoculated in a high-pressure-treated (400 MPa, 17 °C, 10 min) meat model system with added bacteriocins, enterocins A and B, sakacin K, pediocin Ach, and nisin. Nisin was very efficient at inhibiting the recovery of *E. coli*, *Staphylococcus aureus*,

and *Leuconostoc carnosum* during subsequent chilled storage; however, it failed to inhibit the recovery of *Listeria monocytogenes*. The other bacteriocins were effective with *L. monocytogenes*, but not with the other tested bacteria. In high-pressure-treated cooked ham (400 MPa for 6 min at 17 °C), the introduction between slices of alginate films containing enterocin reduced the count of *Listeria monocytogenes* below the level of detection, whereas either high-pressure treatment or active packaging alone had no effect on the bacteria count (Marcos and others 2008a).

The combination of preservation factors or techniques to produce safe, stable, and high-quality food products has been designated the “hurdle concept” by Leistner and Gorris (1995). This concept consists of adding several barriers (hurdles) to bacterial life and growth. For example, in the study of Yuste and others (1998), pressure oscillations (3 cycles of 5 min at 450 MPa at 2 °C), nisin (200 ppm), and weak acidity (pH 5.42 provided by the addition of glucono- $\delta$ -lactone) reduced the psychrotrophic and mesophilic bacterial counts to levels below 2 log CFU/g in mechanically recovered poultry meat during one month of chilled storage. Nisin proved to be more efficient in reducing the indigenous flora of high-pressure-treated poultry meat when used at a weakly acidic pH (Yuste and others 1998, 2002). In fermented sausages, the inherent hurdles (low  $a_w$  and acidity) combined with high pressure (Porto-Fett and others 2010) were used to reduce *Listeria* and *Salmonella* risks. In this study, fermentation and drying of inoculated salami at 7 log CFU/g induced a first reduction of 1.1 to 1.3 log CFU/g for *Listeria monocytogenes* and 4.2 to 4.8 log CFU/g for a *Salmonella* sp. In addition, following pressurization at 600 MPa for 1 to 5 min or at 483 MPa for 5 to 12 min, numbers of *Listeria monocytogenes* and *Salmonella* were reduced by an additional 1.6 to  $\geq 5.0$  log CFU/g and 1.9 to 2.4 log CFU/g, respectively, compared to their levels after fermentation and drying (Porto-Fett and others 2010). The use of lactate, high pressure, and 1 °C refrigeration is a successful combination that could reduce the *Listeria* risk (Aymerich and others 2005; Marcos and others 2008b). In fact, in high-pressure-treated (400 MPa, 10 min at 17 °C) cooked ham inoculated with *Listeria monocytogenes*, a recovery of the bacteria was observed during storage at 6 °C, but no recovery was observed when the ham was formulated with 1.4% of lactate or when it was stored at 1 °C (Aymerich and others 2005).

Storage conditions are thus of great importance for cell recovery after high-pressure treatment. Generally, the lower the storage temperature, the better is the resulting microbial quality, except for certain specific products, such as fermented sausages that benefit from storage at ambient temperature (Jofré and others 2009a).

### High-pressure-assisted sterilization

The high-pressure process has been successfully implemented in some companies as an efficient pasteurization process. Recently, the possibility of using high pressure as a sterilization process has arisen. In fact, spore inactivation can be achieved by combining high pressures with high temperatures (HPHT). This combination is possible when the high-pressure treatment begins at elevated temperatures of between 60 and 90 °C and when using adiabatic compression for rapid heating at temperatures higher than 100 °C (Matser and others 2004). The production of shelf-stable low-acid food (such as meat products) involves the use of severe retorting conditions. HPHT could be used to sterilize these products at lower temperatures and/or with a shorter heat exposure time. Zhu and others (2008) determined the kinetic parameters of the destruction of *Clostridium sporogenes* spores in ground beef treated at 700 to 900 MPa and 80 to 100 °C and confirmed that *Clostridium*

spores can be destroyed more quickly with this method than with conventional processing. For example, the D-value at 100 °C was 16.2 min at atmospheric pressure and decreased to 1.14 min at 800 MPa.

Another way to achieve commercial stability is to use successive high pressure and thermal treatments to induce germination of spores and then destroy them (Moerman 2005; Akhtara and others 2009). In fact, Gould and Sale (1970) and Sale and others (1970) showed that bacterial spores can be germinated at between 300 to 400 MPa and 40 to 50 °C. Spores of *Bacilli* spp. and *Clostridia* spp. were significantly reduced in pork Marengo treated at 400 MPa for 30 min at 50 °C, whereas spores were resistant to the same treatment performed at 20 °C. However, the bacterial spore count can only be reduced to a limited extent (a reduction of 1.6 to 3.8 CFU/g depending on the species) if a single treatment at 400 MPa and 50 °C is performed and cannot provide commercial sterility (Moerman 2005). According to Akhtara and others (2009), a complex and long series and combination of thermal and pressure treatments is necessary to destroy *Clostridium perfringens* spores efficiently (a reduction of 4 log CFU/g of 6 inoculated) in poultry meat. A primary heat treatment at 80 °C for 10 min activates spores for germination. Then, meat is cooled to 55 °C and incubated at 55 °C for 15 min to allow spore germination. Finally, germinated spores are inactivated by pressure-assisted thermal processing at 568 MPa for 10 min at 73 °C.

The main strategy used to increase the efficiency of high pressure for food pasteurization is a combination of heat at approximately 60 °C and high pressure. The addition of several hurdles to microbial growth, like chilled storage, plus the use of specific additives, like bacteriocins or organic acids, allow increased efficiency of the high-pressure treatments. Investigations now explore the possibility of developing pressure-assisted sterilization of meat products either by combining pressure with temperatures higher than 100 °C or by inducing spore germination before destroying them.

### Other Pressure-Assisted Processes Applied to Meat and Meat-Derived Products

In addition to pasteurization, high pressure offers new opportunities in food innovation. Here, the main potential applications of pressure-assisted processes for meat products are described.

#### Freezing and thawing

Because the freezing point of water is reduced at elevated pressure, a food that has been allowed to supercool under pressure (approximately 200 MPa) will undergo rapid freezing once atmospheric pressure is restored. This process, termed “pressure-shift” or “pressure-assisted freezing,” induces the formation of smaller ice crystals and results in less physical damage to food structure (LeBail and others 2002). High-pressure-assisted freezing of pork muscles of 50 mm  $\times$  75 mm  $\times$  11 mm provides a uniform small-size ice crystal distribution in comparison with air-blast freezing and liquid nitrogen freezing (Martino and others 1998). However, a side effect of the process is that the time needed to reach a uniform temperature in the meat piece under pressure is very long. At least 1 h is necessary to attain a uniform temperature of  $-20$  °C in a 50- to 75-mm piece of pork muscle before decompression can take place (Martino and others 1998; Zhu and others 2004). Another side effect is that high pressure, even at a moderate level of approximately 200 MPa, generally causes denaturation of muscle proteins, yielding a cooked appearance of the muscles (Ashie and others 1999; Zhu and others 2004). Zhu and others (2004) showed that pressure-shift freezing causes considerable



modifications in myofibrillar proteins of pork muscle, which leads to an increase in toughness after thawing compared with air-blast frozen muscles.

Similarly, high-pressure treatment of frozen foodstuffs will increase the thawing rate due to the concomitant reduction in the melting temperature of ice and a corresponding increase in the rate of heat flux between the ambient medium and the solid-liquid interface (LeBail and others 2002). High-pressure thawing of cylindrical samples of ground beef with diameters ranging from 55 to 80 mm can be efficiently achieved at 210 to 280 MPa within 30 min and with no quality loss compared to conventional thawing (12 h at 3 °C) (Zhao and others 1998). Okamoto and Suzuki (2002) also found that 200 MPa of pressure provided the best results for the thawing of molded pork meat (70 to 80 mm × 130 to 150 mm, thickness 15 mm) with improved quality in comparison with thawing by running water. In particular, a better retention of water was observed. However, according to Park and others (2006), only thawing under high pressure below 100 MPa can improve the quality of pork compared to atmospheric-pressure thawing in an air-blast freezer at 15 °C. In particular, drip loss is decreased under these relatively low pressures (Okamoto and Suzuki 2002; Park and others 2006). The behavior of frozen food products during pressure-assisted freezing may be affected by the other processing parameters, such as pressure holding time and the initial temperature of the product. For example, the temperature in the product increases more rapidly for samples with lower initial temperatures (Zhao and others 1998), and the high-pressure treatment should be long enough to thoroughly thaw the frozen meat. The optimal thawing time decreases with increasing pressure (Park and others 2006). For example, 60, 48, and 39 minutes are necessary to optimally thaw a pork muscle of 50 mm diameter × 100 mm length at 50, 100, or 150 MPa, respectively (Park and others 2006).

The scale-up of high-pressure/low-temperature processes to an industrial level will require accurate control of the temperature profile in large-scale devices (Urrutia and others 2007).

### Other processes

Application of moderate pressure has been reported to accelerate the diffusion of components into food products and has been used to improve osmotic dehydration of fruits (Rastogi and Niranjana 1998). According to Villacís and others (2008), pressure could also be used to facilitate NaCl diffusion into meat because salt diffusion increases 10-fold under moderate pressure (150 MPa) in comparison with salting at atmospheric pressure.

In addition, Schenkova and others (2007) demonstrated that the Warner-Bratzler shear force of papain-injected beef meat could be reduced by high-pressure treatment at 100 to 300 MPa for 10 min. However, a sensory analysis performed with an untrained sensory panel on papain-injected and pressure-treated (300 MPa, 10 min) cooked meat showed that pressure improves juiciness but does not improve the tenderness in comparison with papain-only treated meat.

Aside from microbial inactivation and meat product texturization, high pressure offers innovation potential for freezing, thawing, or injection processes applied to meat and meat products. Pressure-assisted processes can improve the quality of meat products in comparison with conventionally processed products. However, the scale-up of pressure-assisted processes is complex.

### Conclusions

High-pressure processing is a safe process that can be used to inactivate microorganisms and stabilize their growth during storage

in meat and meat products. Pressure levels higher than 400 MPa are generally necessary to achieve efficient microbial inactivation, depending on the product microbiota and on the meat product itself. Such pressure levels may induce significant changes in the quality attributes of meat and meat products as high pressure has been shown to induce protein denaturation and acceleration of lipid oxidation during subsequent storage. Such modifications can lead to color and texture changes and decreased sensory acceptability. Thus, a number of challenges are evident. The first is to counteract the side effects of pressure by adjusting the process parameters (temperature and pressure cycles) and the formulation of the product itself or its packaging. The combination of hurdles to alter microbial development could also allow for use of a milder pressure treatment. A second challenge is to find ways to take advantage of the modifications induced by high pressure. The potential of high pressure to tenderize meat has been thoroughly studied in the past but has not been applied because meat should be processed at the pre-rigor stage to obtain relevant texture improvement. Currently, many studies deal with the potential use of high pressure for meat-product texturization. There is now a trend to apply high pressure to specific products to ensure texturization and pasteurization simultaneously. High pressure is no longer seen as a simple alternative to conventional pasteurization but as a technique to create innovative meat products.

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