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Original Article

Effect of Fermented Apple Solution on the Textural Properties of Beef Loin Steak

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Summary : The effectiveness of adding fermented apple solution (FAS) along with the mechanical tenderization on mature beef was investigated. The loin steaks from post-breeding Japanese Black Cattle were subjected to shear force examinations after being mechanically tenderized, marinated in FAS and cooked. The mechanically tenderized samples that were treated with wet-heat and dry-heat showed a significant decrease in the values of the breaking strength, when compared to that observed in non-mechanically tenderized samples. The data indicates that the breaking force values of grilled samples that had been treated with FAS were significantly reduced ($p < 0.05$). Compared to untreated samples the samples treated with FAS and vacuum packed, showed a drastic ($p < 0.01$) reduction in breaking strength values. Data suggests that the vacuum packaging provides the opportunity for substances such as FAS to penetrate into the meat tissues more effectively. Top loin steaks of beef treated with FAS retained water much more efficiently than control samples. Results from the sensory evaluation showed that the taste of the samples treated with FAS were extremely good and acceptable. Prospectively, the use of FAS along with the mechanical tenderization treatment and/or packaging steaks with FAS under vacuum condition would contribute considerably to the amelioration of textural properties and tenderization of mature cow's meat. Addressing the application of these studies to meat production may result in the conversion of tough meat into a consumer-acceptable product in the future

Key words : Beef tenderization, Fermented apples, Loin meat, Post-breeding cow meat, Vacuum packaging.

Introduction

The processing of mature cow's meat has become a major interest in meat industries. Tenderness of beef and any red meats is a fundamental factor in its quality and affects the revenue of the meat industry. The most significant difficulty that consumers face today is that the tenderness of the meat cannot easily be determined by visual inspection in the shop.

A survey in Australia found that 77 % of consumers would be prepared to buy more beef if they could be sure that there was an improvement in tenderness compared to their previous purchase (Lawrence *et al*. 2006). Providing tenderized meat, especially beef, seems to be an important factor in meat consumption for consumers of various ages throughout the world, particularly children and elderly people. Some people require meat products that are soft because they find it difficult to bite into and chew tough meats. Babies and elderly people, in particular, fall within the general scope of people with these needs (Muguruma *et al*. 2006). The use of new materials can sometimes help to meet the challenges faced in meat manufacturing. In recent decades, many meat suppliers have started to create various types of soft

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meat by using several techniques such as mechanical tenderization or the use of chemical additives or extracts from fruits and leguminous sources.

The physical disruption of the muscle structure reduces the density of the cooked meat fibers and therefore improves its tenderness (Hokins, 2004). Mechanical tenderization is considered to be a crucial process in meat softening. This process utilizes techniques that break down or diminish the coherent forces in the connective tissues. Meat cuts from mature cows are considered to be the toughest among beef cattle, simply because the tissue is formed by strong connections in the myofibrils of the muscle and in the collagen. In an investigation into the effects of blade tenderization on beef, Pietrasik & Shand (2004) reported that it may be especially necessary for beef products due to the strong collagen connections and differences in the muscle structure. It seems that more rigorous physical treatment of beef is needed in order to produce meat which is as tender as meat from other species. Consumers do not favor tough meat and tenderized meat is always preferred. This makes the tenderization procedure very important in the meat industry, therefore tenderization will elevate the quality of meat produced for commercial purposes because it significantly improves acceptability amongst consumers.

The technologists need to pay particular attention to those various processes that would directly impact the textural quality of meats that are tough (e.g. mechanical processes, additives, heat treatments, vacuum packaging). Two studies were carried out to tenderize meat from older cows and cuts with excessive connective tissue utilizing blade tenderization (BT), hydrodynamic pressure (HDP) and the combination of the two treatments (Liu *et al*. 2006). Generally, cuts from post-breeding and mature cows are used for minced meat, sausages and other products. Therefore, such cuts must necessarily be modified or processed in due course to meet the demands of consumers.

Few studies have been conducted on meat tenderization using plant-derived additives such as papaya, kiwi and ginger. Utilization of fermented apple solution (FAS) as used in the present study as a meat tenderizing material has not yet been fully accepted and the available literature relating to this agent is very limited. Yet, apple has been used by the food industry in the processing of food products and as a food additive. A considerable fraction of these fruits, mainly those not approved for fresh consumption, is industrially processed to produce juices, flavors, and concentrates (Villas-Boâs *et al*. 2003). The effects of adding extract of fermented apples to mature cow's meat had not been investigated until the present study was undertaken.

Therefore, this study aimed to soften tough mature meat through the use of fermented apple solution with wet- and dry-heat treatment after being mechanically tenderized. In addition this research also investigated the effect of vacuum packaging of mature cow's meat once it had been marinated in FAS.

Materials and Methods

Materials

The meat used in this study was loin steak (*Longissimus dorsi* muscle) (Japanese Black Cattle). The animals were fed on finishing diets. The ani-mals used for slaughter were much older than are typically used for retail meat steaks; consequently they had tough myofibrillar structures and connective tissue. The animals' ages were around 6 years old. Each cow had delivered approximately from 3 to 4 calves (obtained from individual animal identification records). The animals were slaughtered at Minami Kyushu Chikusan Kogyo Ltd., Kagoshima, Japan. The meat was vacuum-packed and stored for 4 days in a chilled refrigerator after slaughtering and before being used in this study. The meat grade was defined as A-3 and the pH level of the meat was found to be 5.7.

The samples were designed as slices (4-mm thick) and intentionally divided into two groups. The first set was mechanically tenderized using a mechanical tenderizer (OHMICHI, OMTR-270, Tokyo, Japan) to create clefts in the flesh (Fig. 1), and the second set did not undergo mechanical tenderization. The apple fruits used were cultivated from any tree of the genus *Malus* of the family Rosaceae. Apples were formerly regarded as a species of the pear genus *Pyrus*. Fermented apple solution (FAS) at pH 9.0 was tested as a meat tenderizer. Fermented apple powder was obtained from Kumamoto Flour Milling Company, Kumamoto, Japan.

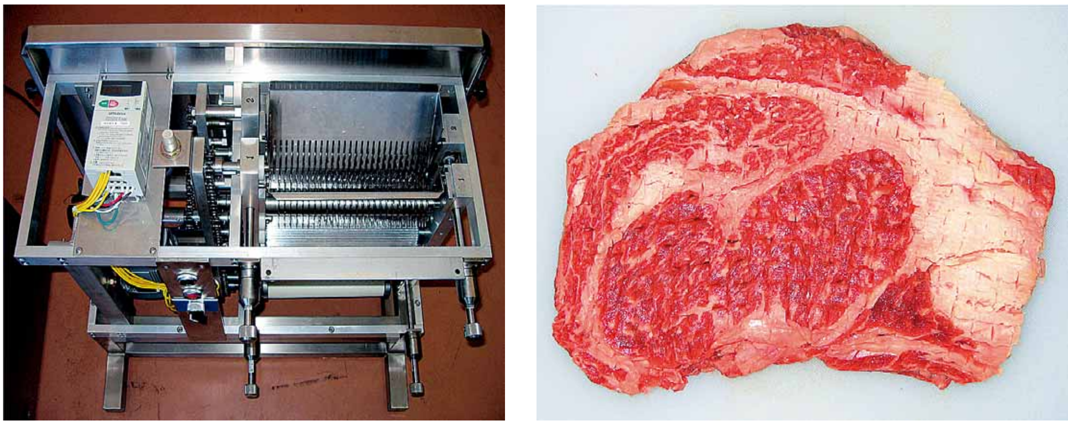

Fig. 1. Image on the left is a typical mechanical tenderizer of meat (OHMICHI, OMTR-270, Tokyo, Japan), image on the right is a photograph of a typical tenderized meat, which was applied in this research.

Treatment of fermented apple solution

Samples in the first set and second set as described in the materials section were also divided into two groups based on the FAS treatments (control and treated). The samples in the first group in each set were marinated in the solution (4 % fermented apple powder) for 3 hours. The ratio of meat to the solution was 2 : 1 (w/v).

The previously mentioned groups were further divided into two subsections; this division was based on the heat treatments. The treated samples after being vacuum packed were cooked using two different heating methods; samples in the first subsection were boiled (wet-heat) at 75°C for 15 min, while the sample slices in the second subsection were grilled (dry-heat) on both sides at 170°C for 5 min in an oven (MRO-NF10, Hitatchi, Tokyo-Japan). In the grilled subsection the steaks were turned once when the temperature and treatment time reached the desired point as previously mentioned.

Effect of vacuum packaging with FAS treat-ment on beef steaks

This research was also concerned with the effect of vacuum packaging on beef tenderization after the addition of FAS which gave the samples a very acceptable flavor. FAS was added to beef slices of the same thickness (4-mm), and then vacuum packed in clear plastic bags using an auto vacuum packer (TOSCI V-306G-II, Tospack, Fukuoka-Japan). The ratio of meat to FAS used in this work was the same as previously mentioned and the samples were kept at refrigeration temperature for one day before testing. Subsequently, the samples were boiled at 75°C for 15 min and were then cooled to room temperature before tenderness evaluation.

The breaking strength of all subsections in both experiments including the controls was determined using a creep meter (Yamaden RE2-33005S, Tokyo, Japan) as used and mentioned in previous studies (Ahhmed *et al*. 2005).

Moisture loss

The method used to cook any meat product contributes considerably to the moisture loss, final weight and size of the meat cut. Moisture loss and weight differences of top loin steaks treated with FAS were determined in this study. The water-retaining capacity (WRC) was determined by comparing the weight of the samples before and after heat treatments. The difference in weight represents the water loss which occurred during both types of heat treatment (water released from meat tissue during the course of boiling or as consequential vapor produced by dry-heat treatment). The cook-ing yield was determined at room temperature by comparing the weight of the sample both before and after the addition of FAS and subsequent heat treatment.

Statistical analysis

This research represents at least five independent experiments and the data was expressed as the means ±SEM ; significant data has also been emphasized in the text. The results of the current research were analyzed with a complete two factorial designs using the additive (FAS) and mechanical tenderization (with or without) as the two factors. The analyses of the data were carried out using ANOVA followed by the Tukey method.

Results and Discussion

The utilization of FAS as a tenderizing substance for mature cow's meat was investigated. The inconsistency in beef tenderness is a major problem in the meat industry. It is estimated that one out of every four choice-grade steaks has a tenderness problem (NCA, 1994 ; Liu *et al*. 2006). Variability in tenderness is a major problem in overall beef quality (Morgan *et al*. 1991 ; Smith *et al*. 1992). Meat tenderness is the most important factor influencing the overall acceptability of beef (Savell *et al.* 1989). Selection decisions at the meat counter are influenced by a variety of factors including price, ease of cutting, quality, workmanship and wholesomeness (AMSA, 2001). Perhaps, ways of cutting the meats are also an influential factor that affecting the consumers decision of purchasing meat.

The texture of tough meats is a very important element in the meat industry ; the softest meat is the most favored amongst consumers especially the elderly. Texture parameters reflected a progressive softening in the meat (Huidobro *et al*. 2003). We suggest that samples in any study relating to methods for softening meats before the addition of any additive and/or any heat treatment must first be subjected to mechanical stabbing. We have reported in a previous study that fracturing the fibers of the flesh improved the absorption properties of the tissues and allowed for greater penetration of the additives (Ahhmed *et al*. 2006). The basic and necessary procedures of meat processing are mechanical tenderization and heat treatments (e.g. stabbing grade, temperature level and treatment time) as well as softening materials. However, the use of these materials is based on the rigidity of cuts in post-harvested meats.

Breaking strength and properties of texture

The samples were subjected to a main test, the textural properties test, often called the share force test. In other words, the breaking strength in all treated samples was evaluated after the heat treatments. In previous studies we have reported that the breaking strength is highly related to the protein complex (Ahhmed *et al*. 2007). In tough meat, the protein complexes which are composed of a group of two or more proteins formed by protein-protein interactions, become rigid over time making the meat tough. The rigidity of the connected proteins together with the connective tissue itself is the main cause of meat toughness. Throughout the lifespan of an animal, the number of cross-links between collagen and fibers increases, this in turn increases collagen heat stability (Lepetit, 2007). Heat treatment also results in the formation of strong complexes in the muscle meat. Cooking is essential for the safe consumption of animal products, since in most human cultures red meat is not eaten raw to improve meat taste and to prevent infection with animal diseases. In recent decades, scientists have devoted considerable resources to the study of all methods of heat treatment. Obuz *et al*. (2004) noted that cooking is another important parameter affecting meat tenderness. Plainly, cooking meat must play a part in the tenderization of meat in addition to its role in reducing the number of microorganisms. Obuz *et al*. (2004) concluded that different cooking methods, heating rates, and size of the samples clearly affected the results obtained. We are in agreement with the recommendations of their study in terms of using small samples for the evaluation of tenderness and how this should be evaluated carefully because tough samples are not solely a result of the usual cooking protocols. We have therefore considered those recommendations as fundamental for the selection of the samples in our study. The samples that we used in our studies were no more than 5 mm in thickness.

Fig. 2 shows the breaking strengths of boiled samples that had been treated with FAS but which had not been mechanically tenderized. The samples were boiled at 75°C for 15 min after being vacuum packed. As a result, the texture of the samples was positively affected by FAS addition. Pietrasik & Shand (2004) reported that mechanical treatments of meat tissue using blade tenderization or tumbling (massaging) are well recognized and accepted techniques in the meat

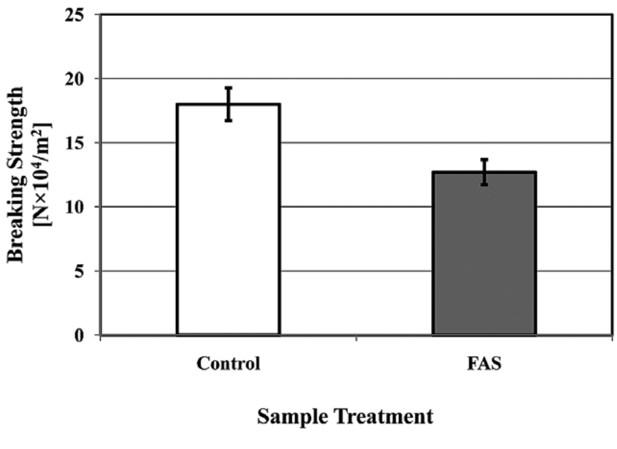

Fig. 2. Effects of fermented apple solution on the breaking strength and texture of mature beef steaks. Samples which had not been mechanically tenderized were boiled at 75°C for 30 min.

industry. Moreover, they reported that the application of blade tenderization prior to injection substantially decreased shear force and TPA hardness, and resulted in a more springy texture when the beef was roasted. Likewise, samples treated with FAS showed a considerable reduction in their breaking strength values. Expressly, the data revealed that the FAS-derived substance used in this study, had improved the tenderness in slices of boiled beef. Clearly, treating samples with FAS resulted in a reduction in the breaking strength of the tissue which was greater than that observed in non-treated samples.

Fig. 3 shows the breaking strengths of boiled samples treated with FAS. The samples were mechanically tenderized before being treated with the FAS-derived substance. The values of the samples treated with FAS and mechanically tenderized were reduced and showed a lower breaking strength than those samples which had not been treated with FAS. Consequently, data revealed that the FAS-derived substance is complementary to any mechanical treat-ment in the production of softer meat. However, the mechanical tenderization with FAS treatment improved the tenderness in boiled slices of beef and prospectively would make a useful contribution to the meat industry.

Furthermore, the breaking strength was measured in grilled samples, which were processed by the same treatments of FAS-derived substance without any mechanical tenderization (Fig. 4). The samples were cooked at 175°C for 5 min on each side and the breaking strength of those samples was immediately evaluated after cooling to room temperature, 25°C. A positive consequence has again been observed in the

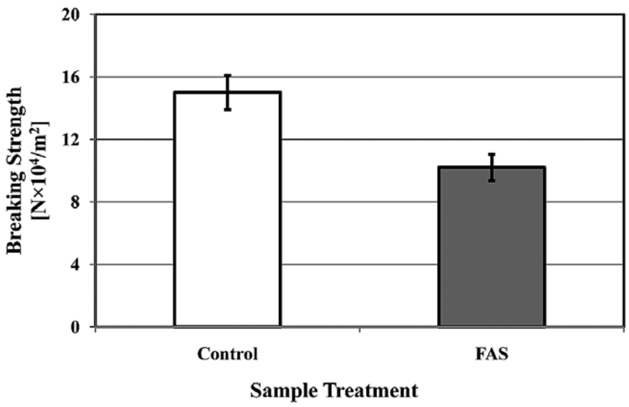

Fig. 3. Effects of mechanical tenderization together with the addition of fermented apple solution on the breaking strength and texture of mature beef steaks. Samples were boiled at 75°C for 30 min. The breaking strength in FAS samples was reduced significantly ($p < 0.05$) when compared to values in control samples.

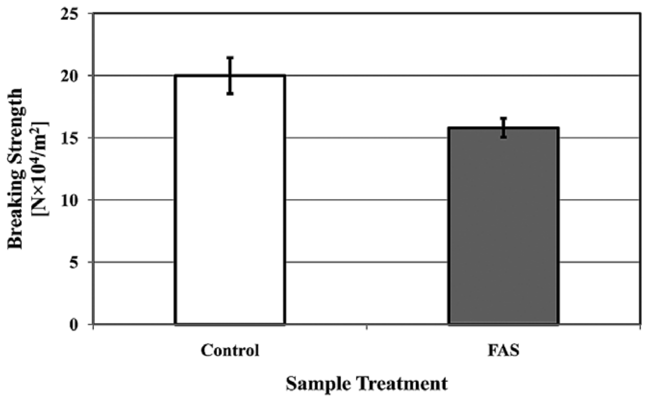

Fig. 4. Effects of treatment with fermented apple solution on the breaking strength and tex-ture of mature beef steaks. Samples which had not been mechanically tenderized were grilled at 170°C for 5 min on each side in a microwave oven. The breaking strength values in FAS samples were significantly reduced ($p < 0.05$) as compared to values in control samples.

texture of the grilled samples, which showed a reduction in the breaking strength. FAS treatment of beef slices was found to be very effective when they cooked using dry-heat. The breaking strength values of samples treated with FAS were significantly reduced ($p < 0.05$). Comparison of wet-heat and dry-heat treatment of samples that had not been mechanically tenderized (Fig. 2 and 4) showed that the FAS-derived substance caused a decrease in the textural hardness of the beef slices.

Fig. 5 shows the values of the breaking strength in grilled samples treated with FAS and mechanically tenderized. The effectiveness of mechanical tenderization along with the addition of FAS was found to be an important method of tenderizing beef slices that are intended and commercially prepared for grilling. As a result, FAS positively affected the texture of mature beef as its breaking strength values were significantly reduced ($p < 0.05$).

Data suggest that the values of breaking strength in most samples treated with FAS and that subjected to two different heat treatments were significantly reduced. Furthermore, in this study samples stabbed prior to the addition of the FAS-derived substance exhibited lower breaking strength values than those samples which had not been subjected to mechanical treatment. Pietrasik & Shand (2005) reported that both the pre-tumbling regime and blade tenderization had a beneficial effect on the textural parameters of roast meats.

Apparently, the slices of beef were positively affected by adding the FAS-derived substance but the

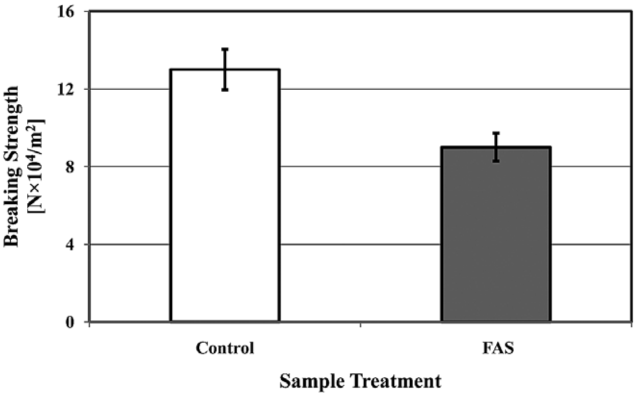

Fig. 5. Effects of mechanical tenderization along with addition of fermented apple solution on breaking strength and texture of mature beef steaks. Samples were grilled at 170°C for 5 min on each side in a microwave oven. The breaking strength values in FAS samples were significantly reduced ($p < 0.05$) when compared to values in control samples.

improvement in the tenderness differed only slightly between samples that were subjected to the two heat treatments. The reasons for this slight difference in texture between samples subjected to the two different heat treatments are as yet unknown. However it can be concluded that the sensitivity of the muscle protein to different types of heat almost certainly plays a part in this phenomenon. Since proteins are less stable in a hot medium, one of the major problems in protein chemistry is the elucidation of a method for maintaining the stability of proteins under conditions of varying temperature and pressure. The actual mechanism of any change occurring during heat and/or pressure treatment is as yet undefined, and it is therefore quite difficult to control the heat-stability of proteins. In a study conducted by Rusman *et al.* (2007) it was found that pressure-heat treatment of up to 200 MPa breaks down the myofibrillar components of the muscle and thus reduces the toughness of the meat but this treatment has little or no effect on the toughness of the connective tissue, because the myofibrillar protein is more sensitive than connective tissue to the effects of pressure.

In the present study, there are two possible mechanisms which could account for the effect of FAS on the tenderization of beef. The first and most likely mechanism is that FAS contains certain enzymes that catalyze the tenderization process. Therefore, we hypothesize that there are certain enzymes present in FAS which can potentially catalyze the connective tissue and/or reduce the force tension in the myofibrillar proteins. Enzymatic proteolytic processes may impact on the myofibrillar structures and induce a considerable decrease in the breaking strength of the beef slices. More likely, we suggest that apples contain several enzymes that contribute to beef tenderness (such as papain in papaya).

As apple is a rich source of many nutrients, those nutrients appear to be retained and also improved in the fermented fruit. Villas-Boâs *et al.* (2003) reported that using various lignocellulose substrates containing low levels of free sugars, previous work has shown that after fermentation with filamentous fungi or yeasts there was an increase in protein content of between 10 and 53 %. The high protein enrichment obtained after apple pomace fermentation probably resulted from the chemical characteristics of the pomace which has high sugar content (Villas-Boâs *et al.* 2003). As fermented apple provides considerable amount of proteins that exist by the act of some microorganisms used the accumulated apple sugar during fermentation process ; there is a high availability of enzymes in such fermented materials. Yang *et al.* (2007) reported that the microorganisms utilize carbohydrates and thereby the proteins are concentrated and since microbes are proteins in nature, ultimately increases the protein content of the product after fermentation. Most likely, extracts of apples may act as enzymatic marinades, which work by breaking down the muscle fiber and connective tissue (muscle protein). These enzymes may provide evidence of the mechanisms responsible for reduc-ing meat hardness.

As many researchers reported that fruits have plenty of enzymes under fresh conditions, we also think that fermented apple solution may be capable in providing such enzymes. Fermented apples could potentially be used as protein supplements (Villas-Boâs *et al.* 2003). Many people consider that extracts of ginger (Naveena & Mendirtta, 2004), kiwi, papaya (Ashie *et al.* 2002), raw pineapple, honeydew and figs all contain protein enzymes (proteases) which may efficiently catalyze the breakdown of proteins if the meat is marinated for an adequate period of time (for few hours or ever for 1 or 2 days). Throughout the world, acid bases including vinegar, wine, citrus juice and tomatoes, are used commercially in many restaurants for the tenderization of meat. Aktas & Kaya (2001) reported that intramuscular connective tissue and sensory properties of beef are influenced as the beef marinated in weak organic acids and salts. Most probably, the bonds between protein bundles in the meat break down and the proteins unwind when meats are exposed to an acidic marinade.

The second probability is that FAS contains some chemical structures that contribute to meat softening, for example alkaline compounds that breach the tight bonds in connective tissues. Thomson & Dobbie (1997) reported that samples treated with sodium chloride for 1 day were significantly tenderer than the controls. Data suggest that FAS would principally reduce the links in myofibrillar proteins and collagen as these are the proteins most responsible for the toughness of animal muscle. Xiong (1997) reported that it is well known that myofibrillar proteins play the most critical role during meat processing because they are responsible for the cohesive structure and firm texture of meat products. The lack of information regarding the effectiveness of FAS on meat proteins makes it difficult to establish it as a softening substance. More studies are necessary to fully understand the role of FAS as a tenderization additive for beef. In many countries, mature beef is considered, in some cases, to be an undesirable meat. Consequently, this study purposes to address the application of these studies to meat production may result in the conversion of tough meat into a consumer-acceptable product in the future.

Effects of vacuum packaging

The second major concern in the current study was the use of vacuum packaging which reduces connecting forces between proteins in beef steak. In a comparison between the untreated samples and samples processed with FAS and vacuum packed, the breaking strength of the treated samples was drastically reduced (Fig. 6). The values of breaking strength in samples that had been marinated for one day in FAS were extremely reduced ($p<0.05$). Breaking strength values in samples marinated in FAS under vacuum pack conditions showed a significant reduction ($p<0.01$) when compared to the values in control samples. Data suggests that the vacuum packaging provides the opportunity for FAS to penetrate into the meat tissues, so that the enzymes and functional elements present in FAS are able to break down the beef proteins. As mentioned previously, FAS was found to be an effective substance for softening meat possibly because of its chemical composition which includes enzymes, proteases or other components such as alkaline moieties. To date, there are two substances used for tenderizing meat, the most popular is bromelain (one of two extracted protease enzymes) which originates from pineapples, and papain which is derived from the leaves of the papaya plant. These are known as proteolytic enzymes because they break down meat protein.

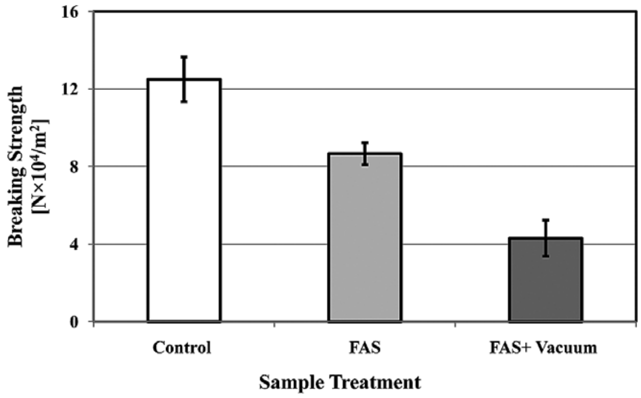

Fig. 6. Effects of mechanical tenderization, the addition of fermented apple solution and vacuum packaging on the breaking strength and texture of mature beef steaks. The samples were boiled at 75°C for 30 min. Values were extremely reduced in samples marinated for one day in FAS ($p<0.05$) and values in samples marinated in FAS under vacuum condition were also significantly reduced ($p<0.01$).

It is very likely that alkaline components in apple play a vital role in the breakdown of myo-fibrillar proteins which leads to the consequent tenderization of beef. Aging meat in solutions which contain alkaline components perhaps plays an important role in meat tenderization, so that marinating samples in such solutions may prove to be an effective method for tenderizing meat.

As fruits including apple contain a considerable amount of calcium, one of the major hypotheses is that apples may increase the calcium content of meat, especially when beef slices are marinated in FAS for a reasonable period of time. It is well known that the calcium in fruits, particularly apples, is responsible for preserving the quality of the fruit during postharvest processes, such as ripening. Ian (2001) reported that calcium plays a role in the prevention of specific disorders such as bitter pit and also contributes to the general quality of the fruit including the degree of firmness of the flesh. By extrapolation of these results we propose that the high concentration of calcium in apples also contributes to its properties in tenderizing beef steak. Ian (2001) also reported on several available methods to ensure that adequate calcium levels are achieved in mature apple fruit, such as the use of both pre-harvest sprays and postharvest dip and drench treatments. These will increase the calcium concentrations in the fruit flesh by, in some cases, up to 30 %. In addition, the content of three important minerals, calcium (Ca), phosphate (P) and potassium (K) also become more concentrated in fermented apple substrate. These minerals increased by 20, 30 and 23 % (w/w), respectively (Villas-Boâs *et al*. 2003). Calcium chloride treatment may improve the quality and consistency of meat in hot-boned bull beef (Thomson & Dobbie, 1997). Many studies have proved that calcium has the potential to reduce the toughness of meat; Gerelt *et al*. (2005) concluded that treatment with 150 mM calcium chloride after osmotic dehydration was sufficient to introduce calcium ions into the meat. In the presence of sufficient calcium, autolysis of calpains and proteolytic degradation of calpastatin which eventually led to a decrease in the rate of calpain and calpastatin activities, clearly seemed to be related to a reduction in the toughness of meat. M-calpain activation was found to be related to an im-provement in meat tenderness (Gerelt *et al*. 2005). Providing additional calcium to muscle has been shown to enhance post mortem proteolysis (Hanson *et al*. 2006). As we marinated the beef samples in FAS for a reasonable period of time, it seemed that steaks had been penetrated by calcium ions. Injection of wether lamb carcass with $CaCl_2$ accelerated the post mortem tenderization process (Polidori *et al*. 2000). Another study conducted by Thomson & Dobbie (1997), provided evidence that calcium chloride treatment pre-rigor increased and post-rigor tended to decrease the shear-force in meat samples. Gerelt *et al*. (2002) concluded that dipping meat in 150 mM calcium chloride solution after dehydration will improve meat tenderness without any detrimental effects on the palatability and other quality traits. Consequently, in the present study, the calcium present in FAS may have penetrated the meat slices during the course of vacuum packaging and thus improved the tenderness of the beef slices making them less hard. As the beef steaks absorbed a reasonable amount of FAS solution which contains calcium ions, this would then increase the activity of the calcium-dependent enzymes. Therefore after a suitable period of time the myofibrillar proteins would be broken down by the activated calcium-dependant enzymes leading to the tenderization of the meat.

Pressure may also play a role in meat tenderization. Probably the pressure that was produced during the vacuum packaging process played a significant role in tenderizing the meat. Rusman *et al*. (2007) noted that a combination of high-pressure and heat treatment seems to be effective for tenderizing tough meat. Meat tenderness is determined by the amount and solubility of the connective tissue, sarcomere shortening during rigor development, and postmortem proteolysis of myofibrillar and myofibrillar-associated proteins (Koohmarie & Geesink. 2006). It has been established that any hydrostatic pressure would definitely damage the architectural fibers in a muscle by stressing the essential connective tissues. Therefore, increasing the pressure is likely to produce more stress which increases the tenderness of the meat. As the texture parameters reflected a progressive softening in the meat (Huidobro *et al*. 2003); in the meat industry the texture of tough meats is regarded as a very important characteristic which determines how the meat is marketed; consumers, particularly the elderly, consider the softest meat to be the most desirable. Results demonstrate that the major effect of increased hydrostatic pressure on muscle fibers which results in tension potentiation, is the com-plete and effective tenderization of meat during processing and it has been shown that the degree of tenderness improves as the pressure continues. This process is an important technique now used in meat industry and the results obtained from vacuum-packed samples treated with FAS would further contribute to the tenderization of mature cow's meat.

Moisture loss

The change in weight of the samples after any heat treatment is thought to be due to water loss. In the current study, we investigated the moisture loss from beef slices cooked using two different methods, dry-heat and wet-heat treatment. These slices were also treated with FAS as described previously. The WRC, expressed as a percentage of the expelled water, increased in heifer meat (Huidobro *et al*. 2003). The potential effect of the FAS substance on the moisture loss and/or its effect on the final weight of the beef slices were also investigated. Control samples also lost water after wet- and dry-heat treatments. The cooking yield was improved by FAS treatment when compared with that of the control samples. Naveena & Mendirtta (2004) also reported that treating buffalo meat with ginger extract improved the cooking yield. Generally salt increases the water retention of meat, but Thomson & Dobbie (1997) also noted that sodium chloride had no effect on cooking yield of 24 or 48 hours post slaughter. We also reported in a previous publication (Ahhmed *et al*. 2007) that the purpose of adding NaCl to chicken and beef sausages was to increase the ability of meat to retain water during cooking. However, in the present study, samples treated with FAS appeared to maintain a satisfactory proportion of their weight (data not shown) which may related to the calcium content of the FAS. Wheeler *et al*. (1991) found that calcium chloride treatment of semi-membranous muscle had no effect on cooking losses.

Moreover, the water released during cooking indicates that some interactions have taken place in beef slices which were marinated with the FAS. Considerable amounts of carotenoids are found in fruits and they are known to attach to a high number of carbon and oxygen atoms. These compounds in particular may facilitate the induction of certain interactions to in the meat proteins, possibly because the proteins have sufficient time to interact with these compounds during the marinating process, which will therefore, aid water retention in the meat. The combination of tenderization with pre- and post-injection tumbling may provide a useful means of improving both textural and water-binding properties (Pietrasik & Shand, 2005). The pH also found to influence the retention of moisture in beef slices. In this study a high pH was found to keep the moisture content of the meat high, as the pH of the FAS was about 9.0. We clearly detected that the WRC is good in samples treated with FAS, since it had a high pH. This concurs with the findings of a previous study conducted by Qiao *et al.* (2001) who found that a higher pH was generally associated with higher moisture content in the meat. The lower percentage of moisture observed in the gel from post-rigor muscle might be related to the complex influences of the lower pH of muscle, leading to shrinkage and fragmentation of myofibrils and a higher protein content in surimi-like pork (SLP) (Kang *et al* . 2007). Hence, in addition to any reaction that occurred during the marinating of beef slices in the FAS substance, the heat treatments also release moisture from meat tissue due to protein shrinkage, and it is therefore suggested that by comparison with control samples, treatment with FAS prevents water loss during cooking.

Sensory evaluation

Each of the samples was also subjected to sensory trait tests. A total of 15 participants of different ages were engaged in this test. Each panelist received a score sheet which was designed according to the Gerelt *et al*. (2002) method. Panelists were asked to mark their preference score in terms of flavor and taste. Seven-point scales ranging from -3 to +3 : very poor (unacceptable product), fairly poor, slightly poor, average, and a slightly above average, fairly good and excellent (most acceptable product) were used for the evaluation. The participants reported that the taste of the samples treated with FAS was influenced by the inherent flavor of the additive. The participants voted positively for the taste of samples treated with FAS and found the taste is extremely good. Furthermore, FAS-treated meat would be preferred by many consumers as is borne out by the results of the organoleptic evaluation (data not shown). In terms of flavor, the palatability of samples treated with FAS was found to be acceptable. Panelists suggested that the use of mechanical tenderization and FAS substance had affected the taste of mature beef; however the effect was positive rather than negative. This was probably because the apertures made by the tenderizer in the flesh of the steaks retained a small volume of these solutions which may have been responsible for the prepotent flavors. We consider this phenomenon to be an advantage rather than disadvantage although it did slightly change the original taste of the meat. Therefore, the decision as to whether this substance should be used by the meat industry is largely dependent on whether the consumer detects a change in the original flavor of the meat.

Conclusions

This research was designed to evaluate the ability of fruit-derived substances to soften the texture of mature beef. From the point of view of quality mechanically stabbing beef slices before adding the tenderizing agent enhances the process of beef steak tenderization. As a result, the breaking strength of boiled and grilled samples was significantly reduced. Using either of the cooking methods together with mechanical tenderization treatment with FAS increased the tenderness of mature beef steaks, reducing the tension force of muscle protein. These findings may partially contribute to a reduction in the toughness of meat. Loin steaks of beef treated with FAS retained water much more efficiently than control samples. The taste of the samples treated FAS was slightly influenced by the taste of apples, and in some cases made them more palatable to the participants in this study. Consecutive treatments using mechanical tenderization and the FAS substance along with vacuum packaging was found to be a very effective technique for softening beef. The vacuum packed samples had the lowest values of breaking strength, which was found to be 60 % less than the breaking strength in the control samples.

In conclusion, this study provides evidence that treating slices of mature beef with FAS may prove to be a useful technique for meat tenderization which could be utilized by the meat industry. It also emphasizes the advantages of using plant-derived substances together with mechanical tenderization to produce meat with an acceptable level of tenderness from mature beef that would otherwise be considered inedible by consumers because of its tough-ness.

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牛ロースステーキのテクスチャー特性に及ぼすリンゴ発酵液の影響

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要　約

経産肥育和牛肉の軟化処理への取り組みを行った. 試料としてはロース肉を用いた. まず, ミートテンダーライザー(OHMICHI, OMTR-270)を用いて物理的な処理を行った. ミートテンダーライザー処理に伴いロース肉の破断強度が低下する傾向が認められた. さらにリンゴ発酵液処理により破断強度はボイル, グリル, いずれにおいても無処理の肉に対して低下することが確認された. またその傾向はミートテンダーライザー処理肉において顕著であった. これはミートテンダーライザー処理肉の方が未処理肉より, ステンレスの刃により肉に筋目を入れることによって, リンゴ発酵溶液が馴染みやすくなる効果が発揮されたものと考えられる. 一方, ドリップロスはリンゴ発酵液処理により改善された. リンゴ発酵液処理によるドリップロスの低減は, 大豆繊維の効果で, より高い保水性が得られ, さらに時間が経過すると線維の間隙が閉じるために, 加熱調理後も肉の水分を保持できることによるものと想定された. さらに, 真空処理を併用することによりそれらの効果が増大することが認められた.

キーワード：牛肉軟化処理, リンゴ発酵液, ロイン肉, 真空パッケージング, 経産牛肉