final report

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HPP concept product development – confirmation of tenderisation and texture assessment

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Abstract

Previous project work (A.MPT.0032) conducted using the large scale 35 L high pressure processing unit at CFNS-Werribee showed that tender meat could be achieved from low-value muscles through the application of high pressure at low temperature. However, several questions arose from this work and areas for further investigation were identified. This progress report outlines preliminary results and methodology used to evaluate objective methods for texture measurement of high pressure processed meat and the cooking methods used for objective and subjective assessment.

Contents

1 Project objectives

- Establish a correlation between objective measurements of texture and sensory preference for texture of HPP treated meat.
  - Objective methods for texture of HPP meat will be evaluated (this report)
  - Cooking methods for objective and subjective assessment will be evaluated (this report)
  - Trained sensory panels will be conducted
- Confirm tenderisation (in replicate) of brisket using identified conditions (400 MPa, 5 min, 4°C) in the 35L unit, with a heated vessel wall
- Investigate the effect of 400 MPa for 5 min at 4°C, without a heated vessel wall, on brisket and other muscle cuts having a medium connective tissue content
- Use the outcomes of the temperature profiling in the 3L HPP unit to investigate the possibility of replicating the processing conditions found in the 0.3L unit at CFNS-Coopers Plains (200MPa, 20 min, 60°C) at a larger scale (i.e. 35L)

2 Success in achieving milestone

This milestone report outlines progress towards the evaluation of objective methods for texture analysis and cooking methods for objective and subjective assessment of high-pressure treated beef samples.

Evaluation of objective methods for texture analysis

Physical structure of a food is key to the perception of its texture. However, forces, deformation and particle properties of the food pieces should be examined to obtain a more complete picture of food texture. Fracture mechanics of food breakdown in the mouth, saliva and time all contribute to texture perception (Hutchings and Lillford, 1988). In cooked meats, these fracture processes for size reduction involve breakdown of the composite, fibrous structure and tensile failure (Lillford, 2001).

The texture of meat depends on the intrinsic mechanical properties and complex arrangement of protein, water and cellular material of which it is made. For example, tenderness and chewiness depend on factors such as the distribution of connective tissue, the elasticity of muscle myofibrils and the integrity of their arrangement (Kerr et al., 2000). Many factors contribute to the basic mechanical properties of meat – cooking, ageing, myofibrillar structure, pH, amount of collagen, and the spatial distribution of connective tissue.

From many rheological studies on cooked meat, Purslow (1985) has suggested that the fracture behaviour of cooked meat is the most important aspect of its mechanical properties in relation to the sensory evaluation of texture. Fracture mechanics determines toughness as the resistance of a material to the propagation of cracks or tears through it in terms of the minimum energy required to propagate the fracture, or by the concentration of stress at the tip of a crack necessary for the rupture to proceed (Purslow, 1985).

Therefore, considering the complexities of the factors affecting meat texture, it is hardly surprising that the perfect instrument for measuring meat tenderness has yet to be devised.

Traditionally, several objective methods for measuring meat tenderness/toughness have been developed – shear force, compression tests, tensile tests, penetrometry, multi-blade shearing tests (Kramer) and bite tests. However, these methods rely on measuring a single parameter and don't imitate the complexity of the chewing motion to breakdown food components or the forces required for deformation and failure.

Of the many methods and instruments developed, perhaps the most widely used and reported is the Warner-Bratzler (WB) shear device. This is usually used to determine the peak force required to shear through a meat sample of fixed cross-sectional area and of known fibre orientation. The conventional WB device uses cylindrical samples and a triangular hole in the shear blade. The modified device (as developed at CSIRO) uses rectangular samples with a square hole in the shear blade (Figure 1). This ensures that the shear blade-sample contact area is constant and makes the shear force deformation curves easier to interpret.

Figure 1: Shear blade for the modified WB shear force device, showing the square hole in the blade.

Texture profile analysis (TPA) is a powerful tool in texture studies and is used for analysing a variety of different foods, e.g. apple, carrot, cheese, meat. TPA simulates the process of mastication and measures the compression force of a probe and the related textural parameters of food during 2 cycles of deformation (Caine et al., 2003). Bourne (1978) adapted the Instron Universal Testing Machine to perform a modified texture profile test similar to that originally developed on the General Foods Texturometer (Friedman et al., 1963).

Isometric tension measurements have been used to follow changes occurring in muscle going into rigor under various conditions (Busch et al., 1972) and also to follow changes taking place in connective tissue during heating (Allain et al., 1978, 1980; Bouton et al., 1978). Due to the complexities of structural interpretation of heat-induced changes in composite systems, this method has not often been used on meat or muscle samples. However, this method could be useful in studying changes produced by heating in meat structural proteins of high-pressure treated muscle.

For this work, assessments of meat texture were made by trialling a number of objective methods – WB shear force (modified method), texture profile analysis (TPA) and hydrothermal isometric tension (HIT) measurements – on two beef muscle with varying connective tissue contents.

3 Methods

Brisket (*M. pectoralis profundus*) and topside (*M. semimembranosus*) muscles from 3 animals were collected from Australian Country Choice (ACC). Animal and carcase information was obtained for each muscle. Samples were cut into muscle portions, approximately 2.5 x 2.5 x 10 cm, and measurements recorded for weight, pH and colour. The samples were vacuum-sealed in polyethylene pouches, and stored at 4°C (1 – 4 h) until treatment. Samples were allocated to either control (no pressure treatment) or pressure treatment, with 5 replicates in each treatment. Samples allocated to each set of pressure treatments came from 1 animal only.

Two sets of pressure conditions identified in previous project work (A.MPT.0013 and A.MPT.0032) to have tenderisation effects were applied to these muscle samples – pressure treatment combined with heat (200 MPa, 60°C, 20 min) or pressure at low temperature (400 MPa, 4°C, 5 min). These treatments were applied on different days, using samples from different animals.

Following pressure treatment, samples were removed from the vacuum pouches, reweighed and pH and colour measurements recorded. Samples were frozen at -20°C until required for texture analysis. For texture assessment (WB shear force and TPA), samples were thawed overnight at 4°C, cooked at 80°C for 30 min in a waterbath and cooled under running tap water. Weights were recorded to measure drip and cook losses. Following overnight storage at 4°C, the cooked samples were cut into subsamples for textural analysis. Details of sample thickness, shape and fibre orientation for samples used for shear force and compression measurements are given in Bouton et al. (1971) and Bouton and Harris (1972). Samples for HIT measurement were thawed at 4°C on the day of testing and analysed uncooked. All textural measurements were made on a Lloyd Instruments LRX Materials testing Machine fitted with a 500N load cell (Lloyd Instruments Ltd., Hampshire UK).

3.1 Warner-Bratzler (WB) shear force

Six subsamples with a rectangular cross section of 15mm wide by 6.7mm deep (1 cm^2 cross-sectional area) were cut from each control and pressure-treated sample, with the fibre orientation parallel to the long axis, and at right angles to the shearing surface (Figure 2). The force required to shear through the clamped subsample with a triangulated 0.64mm thick blade pulled upward at a speed of 100 mm/min at right angles to fibre direction was measured as peak force (N) on a Lloyd Instruments LRX materials testing machine fitted with a 500 N load cell (Lloyd Instruments Ltd., Hampshire, UK) (Figure 3). Parameters measured from shear force deformation curves were peak force (PF), initial yield (IY), and peak force minus initial yield (PFIY). The mean of the subsamples was recorded. An example of the output from a WB deformation curve is shown in Figure 4. Studies on force-deformation curves have shown that treatments influencing predominantly the muscle fibres such as ageing, cooking and myofibrillar contraction affect mainly the IY values, whereas differences between the initial yield and peak force values (PF-IY) reflect changes due to animal age, cooking methods and muscle connective tissue differences (Bouton et al., 1975).

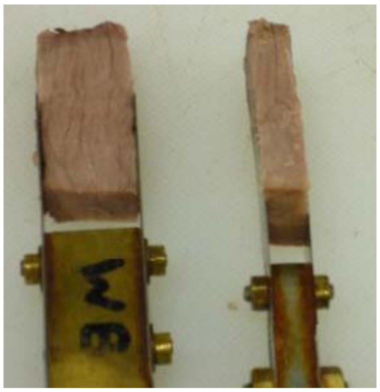

Figure 2: Rectangular sample dimensions for texture analysis using the modified WB shear force device.

Figure 3: Modified WB shear force device.

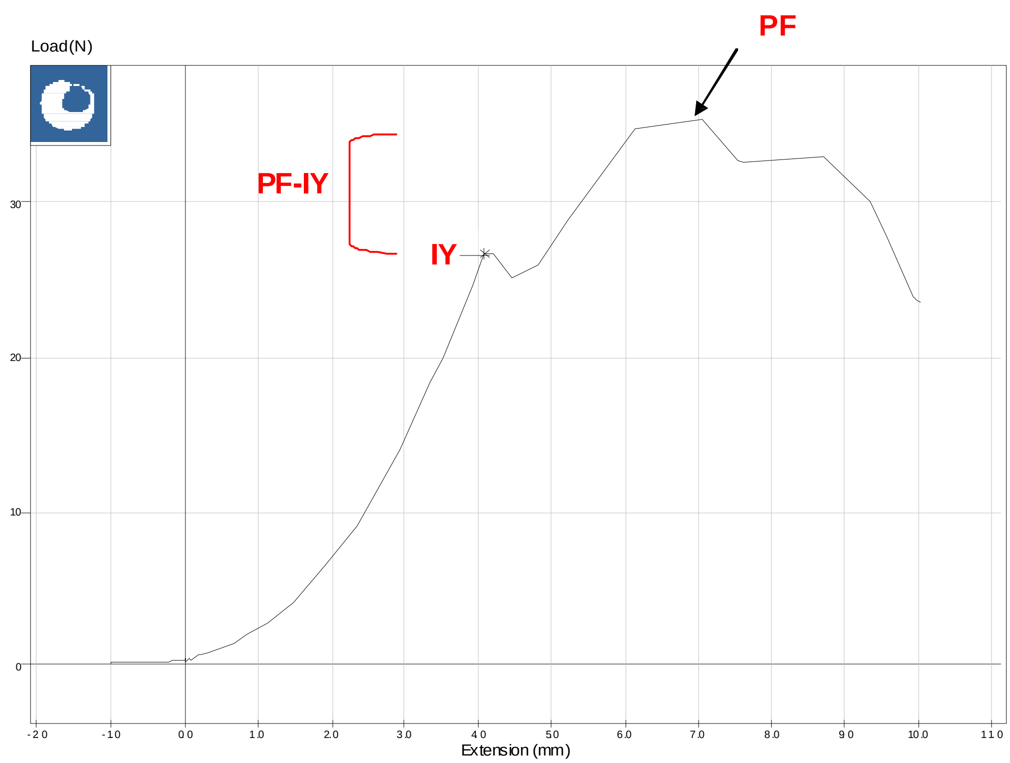

Figure 4: A typical deformation curve from a WB shear force measurement, indicating the peak force (PF), initial yield (IY) and peak force minus initial yield (PF-IY).

3.2 Texture profile analysis (TPA)

One rectangular slice with a thickness of approximately 10 mm was cut from the muscle samples so that the muscle fibre direction was parallel to the largest cut surface. Samples were compressed across the direction of their fibres to 80% of their initial height using a cylindrical plunger with a 6.3 mm diameter and at a speed of 50 mm/min (Lloyd Instruments LRX materials testing machine, 500 N load cell). A minimum of 5 replicates (usually 6-10) was done on each sample, with mean compression recorded in kilograms.

Analysis of the force-time curve leads to seven textural parameters – five measured and two calculated from the measured parameters. A typical TPA deformation curve is shown in Figure 5. The physical description of these attributes has been described by Civille and Szczesniak (1973) as follows:

- Hardness – peak force during the first compression cycle
- Cohesiveness – ratio of the positive force area during the second compression to that during the first compression
- Adhesiveness – negative area for the first compression cycle, representing the work necessary to pull the plunger away from the sample
- Fracturability (brittleness) – force at the first significant break in the curve
- Springiness (elasticity) – rate at which deformed material goes back to its undeformed condition after the deforming force is removed
- Gumminess (compression) – energy required to disintegrate a semi-solid food to a state ready for swallowing – the product of hardness x cohesiveness
- Chewiness – the product of gumminess x springiness (hardness x cohesiveness x springiness)

3.3 Hydrothermal isometric tension (HIT) measurement

Maximum hydrothermal isometric tension (HIT_m) was measured as described by Beilken and Harris (1987). The length and weight of strips of muscle approximately 3 cm long and 2-6 mm wide was recorded and the average cross-sectional area determined assuming a density of 1.04 g/cm^3 for raw meat. The muscle fibres were parallel to the longest length of the sample. Each strip was immersed in distilled water and held tight between two clamps (Figure 6) on a Lloyd Instruments TA Plus Materials Testing Machine fitted with a 500 N load cell (Lloyd Instruments Ltd., Hampshire UK). The distance between the clamps was 10 mm. Slight tension was applied (0.05 N) to the sample and a circulating water bath was used to raise the temperature at a rate of approximately 2°C/min until the force began to decline after reaching a maximum (around 90°C). The apparatus used for this measurement is shown in Figure 7.

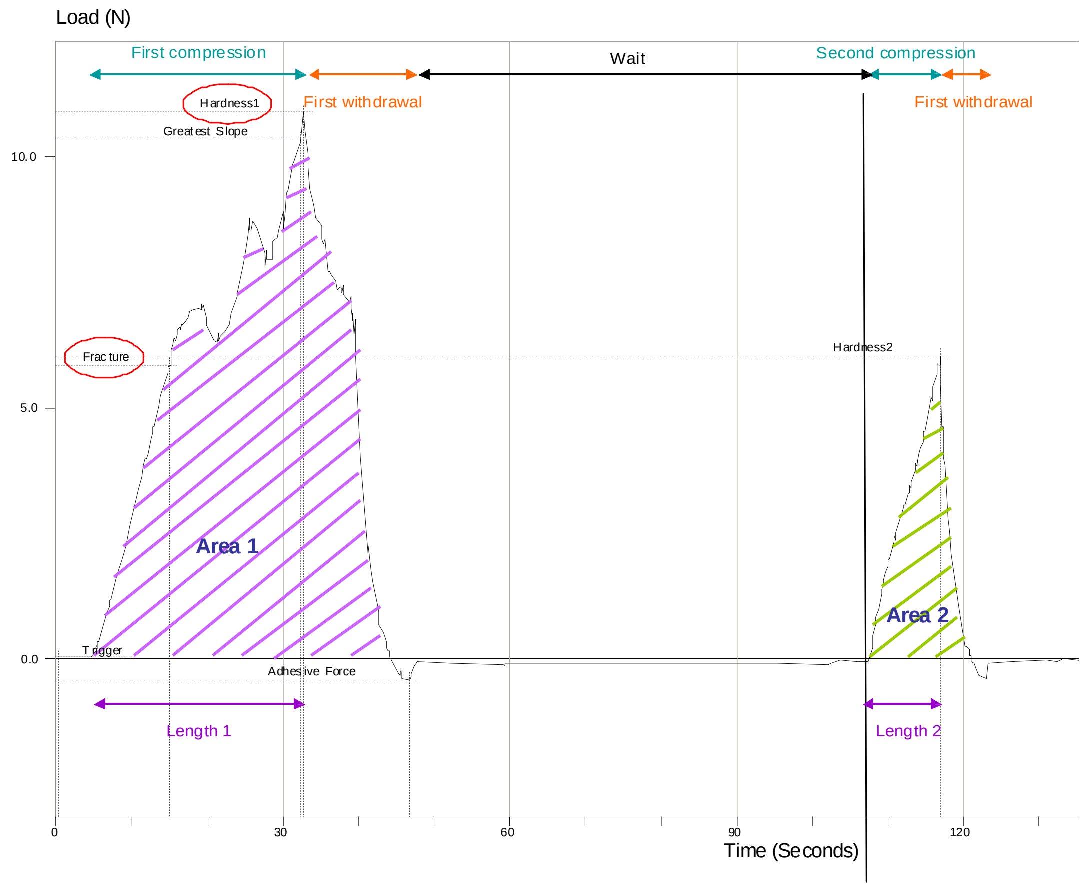

Figure 5: A generalised TPA deformation curve indicating the different parameters that can be measured.

Figure 6: Meat sample held between serrated jaws for HIT measurement.

Figure 7: Apparatus and set-up used for HIT measurement.

4 Results

4.1 Warner-Bratzler (WB) shear force

The effect of pressure combined with heat on the texture of brisket and topside, as measured by WB shear force, is shown in Figure 8. Although not significant, the trend appears to be tenderisation of brisket muscle but a toughening of topside. However, the increase in WB shear force is marginal for topside. It also should be noted that these treatments were applied to samples from only 1 animal.

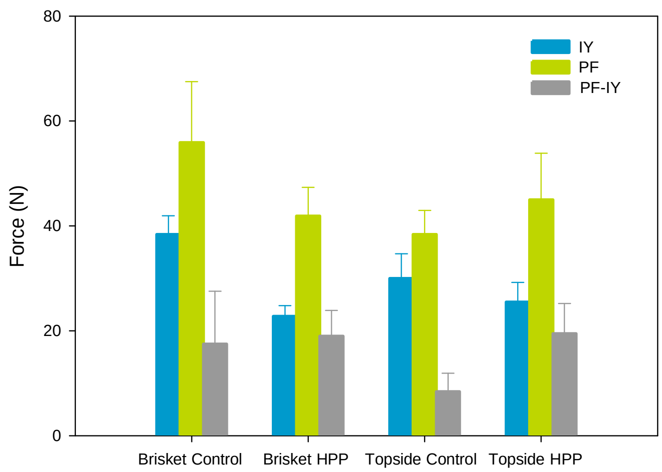

Figure 8: WB shear force measurements of untreated (control) and pressure treated (200 MPa, 60°C, 20 min, HPP) brisket and topside beef samples. IY – initial yield; PF – peak force; PF-IY – peak force minus initial yield. (Mean ± SD, n=5)

There was a greater impact on the myofibrillar component (IY) of the pressure-heat-treated brisket compared to the untreated brisket, with little effect on the connective tissue component (PF-IY). In contrast, the application of pressure combined with temperature to topside had minimal effect on the myofibrillar component but an effect on the collagen component (increased force) (Figure 8).

When pressure was applied at low temperature (400 MPa, 4°C, 5 mins), no change in the texture of brisket was observed compared to the untreated sample, whereas a tenderisation effect was evident for topside, although not significant (Figure 9). As for the pressure-heat treatment, pressure at low temperature was applied to samples from only 1 animal.

It has previously been shown that shear force values were more influenced by the muscle fibre properties than by connective tissue properties of muscle samples (Bouton and Harris, 1972; Cross et al., 1973). Shear force values have also been shown to correlate poorly with subjective assessment of tenderness when there are large differences in connective tissue strength (Bouton et al., 1973).

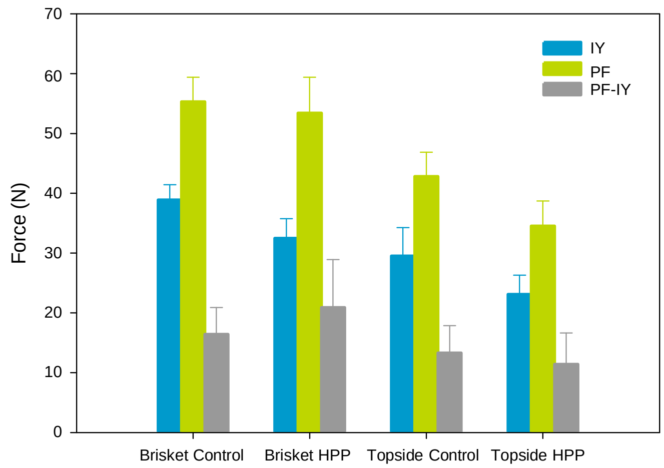

Figure 9: WB shear force measurements of untreated (control) and pressure treated (400 MPa, 4°C, 5 min, HPP) brisket and topside beef samples. IY – initial yield; PF – peak force; PF-IY – peak force minus initial yield. (Mean ± SD, n=5)

4.2 Texture profile analysis (TPA)

The seven parameters measured by TPA of brisket and topside treated with combined pressure and temperature are shown in Figure 10. There is no effect on the hardness of topside whereas a tenderising effect on brisket is evident.

There was no effect on the hardness of either brisket or topside when treated with pressure at low temperature (400 MPa, 4°C, 5 min) (Figure 11).

Careful scrutiny of the other TPA parameters needs to occur to see if there are any relationships.

5 Conclusions

The sensitivity of the mechanical devices (WB or TPA) to changes in the texture of high pressure-treated meat samples could be dependent on whether the changes were a result of changes in either muscle fibre properties or connective tissue properties.

Results from previous work reported in the literature needs to be considered when deciding on a method for texture assessment of high-pressure treated muscle. For example, Bouton and

Harris (1972) showed that shear force values were more influenced by the muscle fibre properties than by connective tissue properties of muscle. Ratcliff et al. (1977) used a combination of objective methods (WB shear force, compression and adhesion) to measure texture of high-pressure treated beef muscles and found that compression (TPA) was the best parameter to measure tenderness even though WB shear force showed the largest effect of pressure treatment.

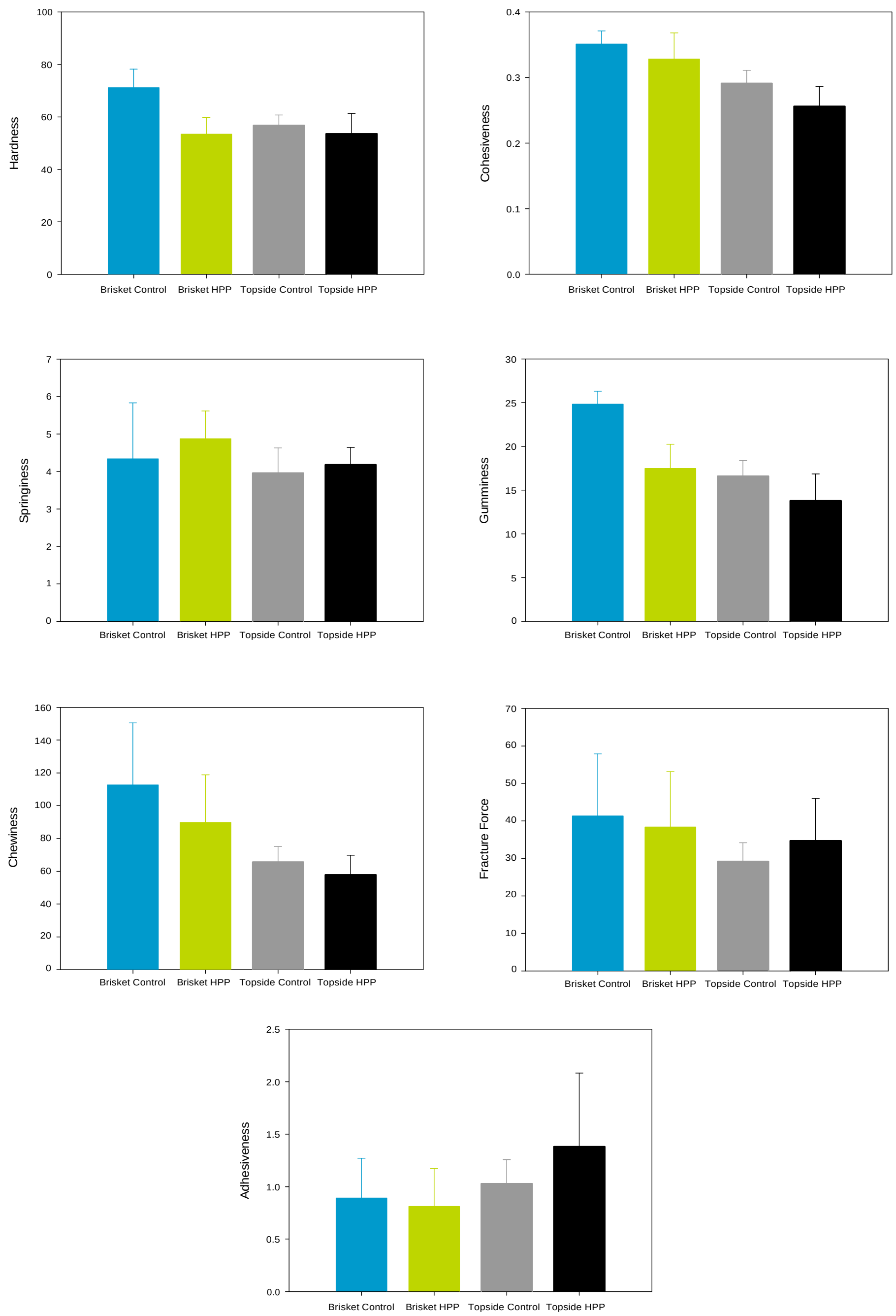

Figure 10: TPA of untreated (control) and pressure treated (200 MPa, 60°C, 20 min, HPP) brisket and topside beef muscle. Mean ± SD, n=5

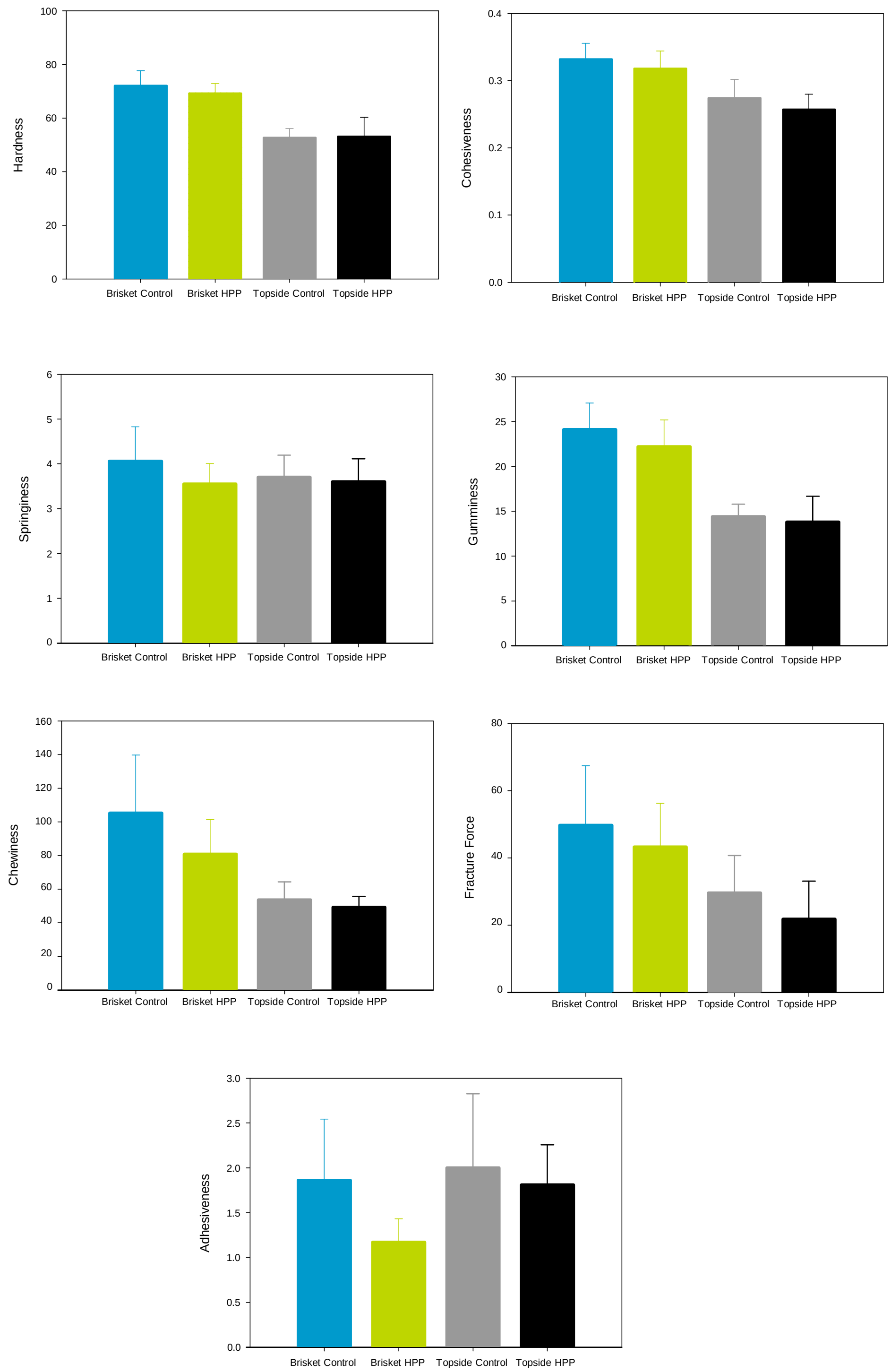

Figure 11: TPA of untreated (control) and pressure treated (400 MPa, 4°C, 5 min, HPP) brisket and topside beef muscle. Mean ± SD, n=5

5.1 Hydrothermal isometric tension (HIT) measurement

Preliminary investigation of the HIT measurement on brisket muscle has indicated differences in the force-time deformation curves for untreated (control) and pressure-treated (200 MPa, 60°C, 20 min) samples (Figure 12). From these initial results, it can be seen that higher contraction forces were generated for the pressure-treated sample. Further examination of this technique is warranted which might convey that some of the detail in the HIT curves could be related to the modification of muscle proteins due high pressure treatment.

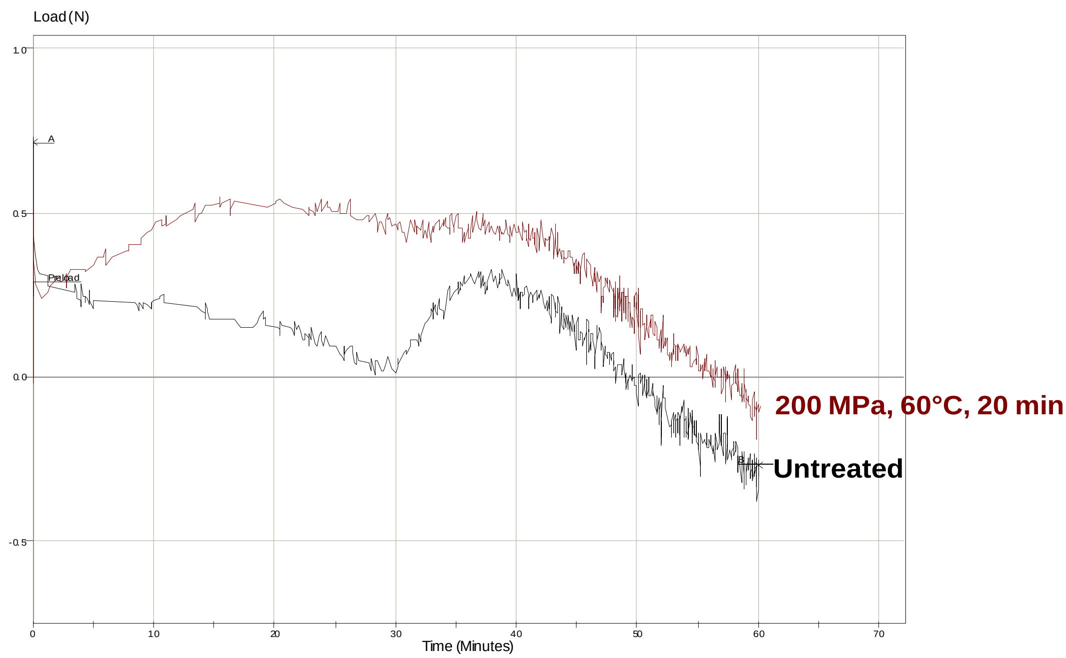

Figure 12: Force-time deformation curve from HIT measurement of untreated and pressure treated (200 MPa, 60°C, 20 min) brisket samples.

5.2 Evaluation of cooking methods

As meat is usually cooked before it is eaten, there have been many investigations into the effects of cooking conditions (temperature, duration) on meat texture. The effects of cooking have been summarised as producing a softening of the connective tissue by conversion of the collagen to gelatin, accompanied by a toughening of the meat fibres due to heat coagulation of the myofibrillar proteins. Harris and Shorthose (1988) suggested a standard procedure for cooking meat samples for texture assessment – samples of a known size in polyethylene bags totally immersed in water controlled at 80°C for a set time.

Methods

Topside (*M. semimembranosus*) muscle was collected from Australian Country Choice (ACC) from one animal. Animal and carcase information was obtained. Samples were cut into muscle portions, approximately 2.5 x 2.5 x 10 cm, and measurements recorded for weight, pH and colour. The samples were vacuum-sealed in polyethylene pouches, and stored at 4°C (1 - 2 h) until processing. Samples were allocated to either control (no pressure treatment) or pressure treatment (200 MPa, 60°C, 20 min), with 5 replicates in each treatment.

Following pressure treatment, samples were removed from the vacuum pouches, reweighed and pH and colour measurements recorded. Samples were frozen at -20°C until required for texture analysis by the WB shear force method. Samples allocated to the different cooking protocols were prepared as outlined below.

Silex® Cooking Protocol

Meat samples were cooked from a frozen state on a preheated (200°C) electric grill (Silex® T10 kitchen genius, Germany) for 13 minutes, turning after 6 minutes. The internal temperature of the samples was measured using a hand-held temperature probe to ensure a minimum temperature of 72°C had been reached. Samples were cooled and stored overnight at 4°C for WB shear force measurement the following day.

Waterbath Cooking Protocol

Frozen samples were thawed overnight at 4°C and cooked in a waterbath at 80°C for 30 minutes. Samples were cooled in running tap water and stored overnight at 4°C for WB shear force measurement the following day.

5.3 Results

As the protocols for the cooking methods differed as to whether the samples were cooked from thawed or frozen, it is only possible to compare the controls and high pressure processed samples from a similar cooking treatment.

There was a reduction in the PF value of the pressure-treated topside sample following cooking in a waterbath, indicating a tenderisation effect. This appears to be mainly due to a reduction in the myofibrillar component (IY) (Figure 13). However, when the topside samples were cooked on the Silex® grill, there was no change in the PF value between the untreated control samples and the pressure-treated sample. There was a decrease in the IY value but the opposite effect on the PF-IY, indicating a different effect on the muscle components compared to the waterbath cooked samples.

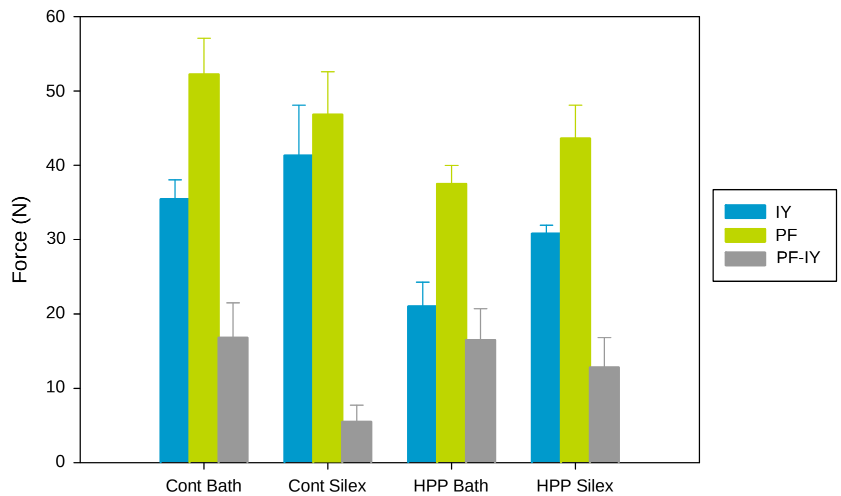

Figure 12: WB shear force values of untreated (Cont) and pressure-treated (200 MPa, 60°C, 20 min, HPP) topside after cooking by either the Silex® protocol (Silex) or waterbath method (Bath). Mean ± SD, n=5.

6 Overall progress of the project

In this work, two methods for assessing meat texture of high-pressure treated beef have been compared – Warner-Bratzler (WB) shear force and texture profile analysis (TPA). Preliminary investigation of hydrothermal isometric tension (HIT) measurement has indicated changes produced by heating in muscle proteins of high-pressure treated muscle. This method will continue to be investigated,

Trials to confirm tenderisation of brisket and topside using previously identified processing conditions in the 35 L high pressure unit are planned in Werribee from June 27 – July 1, 2011. Due to engineering issues with the 0.3 L high pressure unit at CFNS-Coopers Plains, sensory assessment using trained panels has been rescheduled to August, 2011.

7 Recommendations

- From results of WB shear force and TPA measurements and previous results in the literature, further discussion is required on preferred texture method for assessment of high-pressure processed meat. Measurements in this report were carried out on samples from only 1 animal.
- Continue investigation of HIT measurements and analyse results to determine if changes in muscle proteins due to high pressure treatment can be detected.
- Standardise the cooking process for texture measurement of high-pressure processed muscle using the Silex® grill protocol. Silex® grills have been used for all sensory research into meat products by a company commissioned by MLA and is the industry standard for consumer panels (MSA).

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