

Optimisation of sous vide processing to enhance the quality of beef short ribs

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Research Article

Keywords: beef short ribs, sous vide, process optimisation, quality, tenderness, colour

Posted Date: March 17th, 2022

DOI: <https://doi.org/10.21203/rs.3.rs-1455971/v1>

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Abstract

The aim of this research was to study the effect of sous vide temperature (60, 65, and 70°C) and time (12, 24, and 36 h) combination on the quality of beef short ribs namely colour, cooking loss (%), shrinkage (%), soluble collagen (%), myofibrillar fragmentation index (MFI), Warner Bratzler Shear force (WBSF), and Texture Profile Analysis (TPA) parameters. Aluminium and polyethylene, ethylene vinyl alcohol, and polyamide plastic laminate pouch was recommended for sous vide processing due to no detrimental effect on colour owing to its low oxygen permeability. The results showed a significant ($p < 0.05$) increase in shrinkage in volume (%), cooking loss (%), and the proportion of soluble collagen (%) and MFI when sous vide temperature and the processing time were increased to 70°C and 36 h, respectively. WBSF and the TPA hardness value were significantly lower ($p < 0.05$) at 70°C than at 60°C both for 36 hours due to higher level of solubilisation of collagen and myofibrillar fragmentation by prolonged heating in the moist in-pack environment and overpressure created by saturated steam. As the use of a laminate pouch reduced the oxidation of red myoglobin to brown metmyoglobin, the effect of sous vide temperature and time on International Commission on Illumination (CIE) hue angle and Chroma was not significant. Based on the data obtained, process optimisation using third order regression was conducted and the results revealed that optimum sous vide processing temperature and time combination for beef short ribs was 60°C and 34.06 h.

1 Introduction

Sous vide (French for “under vacuum”) processing has been used in home cooking and by chefs. Sous vide method applies heating at controlled temperature and time regimes inside heat-stable vacuumed plastic pouches (Schellekens, 1996). The precise temperature and time control has benefits of impeccable reproducibility; good control over degree of doneness, no detrimental effect on colour and improved tenderness of tough lower value meat cuts (Baldwin, 2012). Vacuum sealing the meat in a bag means that heat transfer from the water (or steam) to the food can be very efficient and it ensures the shelf life and safety of the cooked food by eliminating the risk of contamination during storage. It also limits off-flavour development by reducing oxidation or evaporative losses of flavour volatiles and moisture loss during cooking. Packaging in low oxygen permeable films also reduces aerobic bacterial growth, which further increases the shelf life of the food (Church & Parsons, 2000; Gonzalez-Fandos et al., 2004; Lassen et al., 2002; Stea et al., 2007).

During sous vide processing, the heat applied can be sufficient to denature some of the proteins, which changes the structure and properties of meat proteins ultimately affecting meat quality. The myofibrillar proteins (mostly myosin and actin) shrink transversely and longitudinally, the connective tissue proteins (mostly collagen) contract and solubilize, while the sarcoplasmic proteins aggregate and gel (Baldwin, 2012). The extent and the type of denaturation, contraction, and expansion of meat proteins mainly depend on the rate of heat transfer, temperature, and time (Baldwin, 2012; Dominguez-Hernandez et al., 2018; Tornberg, 2005). The fastest changes in the meat proteins begin at temperature range between 35 and 40°C with shrinkage of myofibrils, at a linear rate up to 80°C. The aggregation and gelation of sarcoplasmic proteins starts around 40°C and finishes at around 60°C.

During heating at temperature of 60–70°C, meat tenderness increases due to solubilisation of collagen and reduction in inter-fibre adhesion (Baldwin, 2012; Roldán et al., 2013). The collagen fibres begin to shrink at temperature around 60°C and at above 65°C, they contract more intensely destroying the triple-stranded helix structure, which transforms the collagen fibres into random coils that are soluble in water, which further enhances tenderness. The denaturation of collagen increases exponentially with an increase in temperature (Baldwin, 2012). Low temperature long time sous vide cooking, with temperature range from 58 to 75°C and processing time in the range of 6.5 up to 24 hours, have been reported to solubilize collagen and can tenderise tough meat (Alahakoon et al., 2018; Bhat et al., 2020; Bouton & Harris, 1981; Christensen et al., 2013; García-Segovia et al., 2007; James Bryony & Yang Seo, 2012; Vaudagna et al., 2002).

Out of the 20 myofibrillar proteins present in meat fibres, actin (forming the thin filaments of myofibrils) and myosin (forming the thick filaments of myofibrils) are the two predominant fibrous proteins, comprising about 65–70% of muscle fibres and holding 70–75% of the water in the meat muscle. The water holding capacity (WHC) of meat during cooking is related to the shrinking and swelling of myofibrils, which depends on the heating temperature. Muscle fibres shrink at 35–40°C, with the extent of shrinkage increasing almost linearly up to 80°C (Baldwin, 2012; Bertram et al., 2006; Bertram et al., 2002). At temperatures between 40 and 60°C, the muscle fibres shrink transversely, which increases the inter-myofibrillar gaps. Above 60–65°C, the muscle fibres shrink longitudinally, which reduces their volume and ultimately WHC and hence causes substantial water loss. The extent of longitudinal contraction and the percentage of cooking loss in meat increases with increasing temperature (Baldwin, 2012).

Meat colour is affected by the rate of change in temperature and time at a particular temperature. In general the faster the heating rate is, the redder the meat colour will be and the longer the holding time occurs, the paler the meat colour becomes. This is because during heating the oxymyoglobin (red) and deoxymyoglobin (purple) is oxidised to metmyoglobin (brown). The ratio of oxymyoglobin, deoxymyoglobin, and metmyoglobin determines the final colour of cooked meat (Baldwin, 2012).

Prolonged cooking (e.g., braising) has been used to make tough meat more palatable. However tough meat such as pork shoulder and beef chuck, which has large amount of connective tissue, can take up to 10–12 h at 80°C or 1–2 days at 55–60°C to become fork-tender (Baldwin, 2012) and the process is not commercially viable on a large scale. So far, the effect of sous vide on bone-in meat like short ribs, which are tough and contain high amount of connective tissue and fat, has received little academic interest. The heterogeneous level of connective tissue varies dramatically according to location on the carcass and among animals. Furthermore, how the degree of meat attachment to the bone changes during cooking needs to be considered. Thus, in order to facilitate the commercial production of sous vide short ribs and to add value to this meat cut, the effect of sous vide on quality parameters of short ribs need to be studied and optimum sous vide temperature and time should be determined. Therefore, the aim of this study was to investigate the effect of temperature and time during sous vide processing on the quality properties of beef short ribs namely colour, cooking loss (%), shrinkage (%), soluble collagen (%), myofibrillar fragmentation index (MFI), Warner Bratzler Shear force (WBSF), and Texture Profile Analysis (TPA) parameters. In addition, the temperature and time to obtain optimal quality properties of beef short ribs was determined.

2 Material And Methods

2.1 Meat samples

Beef short ribs (*Transversus thoracis* muscle) from both left and right side were obtained from five steers (15 months old, Angus breed) raised in the same pasture-based farm. Pre-rigor short ribs were stored at 15°C for 48 h until rigor, vacuum packed, and frozen to -18°C. Before commencing the experiments, each short ribs consisting of five bones were cut longitudinally to 3 pieces (~ 8 x 8 x 6 cm³ pieces) and thawed at 4°C for 16 h. Small size bones were discarded.

The study used a 3x3 full factorial design with sous vide temperatures of 60, 65, and 70°C and sous vide times of 12, 24, and 36 h. To allocate the process variables, sous vide time was allocated to randomized short ribs from bone 1 to 3 (from top to bottom) and sous vide temperature (60, 65 and 70°C) and a control were assigned using Latin square design and blocked by replicate. The number of replicates for each treatment combination was 10.

2.2 Sous vide treatment

A pre-study investigating the effect of different packaging materials on meat appearance after sous vide treatment was carried out. Thermo-resistant vacuumable plastic polybags pouch and laminate Excel Foil pouches (Contour Sales Packaging Systems, Tauranga, New Zealand) were used. The polybag pouches had a thickness of 70 µm, oxygen permeability was 50 cc/m²/day at 23°C, carbon dioxide permeability was 200 cc/m²/day at 23°C, nitrogen permeability was 12 cc/m²/day at 23°C, and the water vapour transmission rate (WVTR) was 8 g/m²/day at 38 °C and a relative humidity of (RH) 90%. The Excel foil pouch was a laminated aluminium and three polymer layer film (polyethylene, ethylene vinyl alcohol and polyamide) laminate of glued with food grade adhesives that had a thickness of 79 µm, an oxygen permeability of ≤ 0.1 cc/m²/day at 23°C, 65% RH, and a WVTR of ≤ 0.1 g/m²/day at 23°C.

Short ribs cuts were vacuum packed (0 bar) in either polybags or laminate bags and sealed by the vacuum packaging machine (Audionvac VMS 153, Derby, UK). The vacuum-packed short ribs were heated by submerging them a water bath (Polyscience Illinois, USA) at the required temperature (60, 65, and 70°C) for 12, 24, or 36 h. The temperature-time profile of the water bath and inside the packaging was monitored using i-button (Thermochron, Deventer, The Netherlands). At the completion of sous vide treatment, the packed meat was immediately submerged in an ice-water bath for 1 hours. The pack was then opened and the meat temperature was held at 4°C for up to 24 h. During this period, shrinkage in volume (% change in volume before and after sous vide), Commission Internationale d'Eclairage (CIE) $L^*a^*b^*$ colour parameters, cooking loss, and texture parameters were measured. The remaining samples were stored at -80°C for further analysis.

2.3 Determination of meat colour

CIE - L^* (white- black), a^* (red-green) and b^* (yellow-blue) values were measured using Hunter Lab Mini Scan XE Plus (Hunter Associates Laboratory Inc., Reston, VA, USA) with light source set at D-65 and observer set to 10°. The colorimeter was calibrated on a black tile and then a white enamel tile using Illuminant D-65 as directed by the manufacturer's specifications. The positive values of L^* , a^* , and b^* correspond to the higher degree of lightness, redness, and yellowness of meat, respectively. Five measurements were taken for each sample and the average values were reported. Similarly, Chroma ($C^* = \sqrt{a^{*2} + b^{*2}}$), and hue angle ($h = \tan^{-1} \frac{b^*}{a^*}$), $\Delta E^* = \sqrt{\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}}$, and $\Delta H^* = \sqrt{\Delta E^{*2} - \Delta L^{*2} + \Delta C^2}$ where ΔL^* , Δa^* , Δb^* , and ΔC are difference in L^* , a^* , b^* and C^* between the sous vide treated meat and raw meat.

2.4 Determination of cooking loss (%)

Cooking loss was measured as a percentage of loss in weight before and after sous vide treatment as described in Eq. (1).

$$\text{Cooking loss(\%)} = \frac{(\text{weight before sous vide} - \text{weight after sous vide})}{\text{weight before sous vide}} \times 100$$

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2.5 Determination of texture parameters

Two texture methods, WBSF and TPA, were used to assess texture using a TA XT-2i Texture Analyser (Stable Micro Systems Ltd., Surrey, UK). Three cylindrical cores (2.0 cm length; 1.27 cm diameter) from the centre of each sous vide processed short ribs were cut using a core borer of 1.27 cm diameter. WBSF was measured by shearing the cylinders perpendicular to the direction of muscle fibres using a HDP/BS Warner Bratzler blade with test speed of 10mm/sec. The peak shear force (N) during shearing was reported as WBSF.

For TPA measurement, each sous vide processed short ribs was cut parallel to the muscle fibre direction to form five 1 cm³ cubes. The short rib cubes were compressed axially in a 2-cycle sequence by a flat cylindrical plunger (P/50) perpendicular to the direction of muscle fibre. First, they were compressed up to a 50% of their original height at a crosshead speed of 2 mm.s⁻¹. The plunger then recoiled to the original position and the meat cubes were compressed again to 100% height at the same speed (Alahakoon et al., 2018). The TPA parameters determined from the force deformation curve were hardness, adhesiveness, cohesiveness, springiness, gumminess, chewiness, and resilience. Hardness was recorded on the first compression cycle of the probe. Adhesiveness was the area, in arbitrary instrumental units, of the negative peak beneath the base line of the profile, and represents the work necessary to pull the plunger from the sample. Cohesiveness was defined as a ratio of the area, in arbitrary units, under the second peak and the area under the first peak (Nishinari & Fang, 2018).

2.6 Determination of soluble collagen

The amount of soluble collagen after sous vide processing was determined using a modified procedure from Hill (1966), in which 4g of meat was heated at 77°C in 12 ml of 25% strength Ringer's solution for 70 min, with shaking at 10 min interval. The samples were then centrifuged (2300 ×g at 4°C for 30 min) and the supernatant containing the soluble collagen was collected. Ringer's solution (8 ml) was added to the meat residue and the mixture was centrifuged (2300 ×g at 4°C for 30 min) and the supernatant was collected. The pooled collected supernatant was hydrolysed in 30 ml H₂SO₄ (7N) for 18 h at 105 ± 1°C and the hydroxyproline concentration and the amount of soluble collagen were determined (Kolar, 1990). To determine the total collagen, 4g of minced sous vide processed meat was hydrolysed in H₂SO₄ (7N) for 18 h at 105 ± 1°C and the hydroxyproline content determined. This was converted to collagen content by multiplying the hydroxyproline content by 7.25 for total collagen (Goll et al., 1963) and 7.52 for soluble collagen (Cross et al., 1973). The proportion of soluble collagen was expressed as a percentage of total collagen.

2.7 Determination of myofibrillar fragmentation index (MFI)

MFI was determined according to a modified method of Aroeira et al. (2020). First, 1 g thin slices of lean meat were mixed in 40 mL cold (4°C) buffer solution (100 mM- potassium chloride, 1mM, 20 mM- potassium phosphate, 1mM- magnesium chloride, 1 mM- sodium azide and 1 mM- ethylenediaminetetraacetic acid disodium salt) and homogenised using an ultra-turrax (IKA® Works (Asia) Sdn Bhd, Selangor, Malaysia) at 15000 rpm for 30 s for three runs. The resulting suspension was centrifuge at 2300 ×g for 30 min and the supernatant was discarded. The pellet was re-suspended in ice-cold buffer (40 mL) and re-centrifuged (2300 ×g, 30 min). The resulting pellet was suspended in buffer (20 mL) and homogenised. Finally, filtrate from a metal sieve (18 mesh) was collected.

The protein concentration of the filtrate was determined using the Lowry method (Wrolstad et al., 2005) after diluting the filtrate to a final protein concentration of 0.5 ± 0.05 mg/mL with buffer. The absorbance of the diluted protein suspensions was measured immediately at 540 nm with a spectrophotometer (Speccord 50 plus, Analytik Jena GmbH, Jena, Germany). The mean of the triplicate absorbance readings was multiplied by 200 to expand the absorbance units to obtain the MFI (Culler et al., 1978).

2.8 Visual grading for the degree of meat attachment to the bone

The degree attachment of the meat to the bone was visually ranked on 1–5 ordinal scale, with 1 being meat attached completely to the bone and 5 being the meat completely separated from the bone.

2.9 Data analysis

The effect of sous vide processing parameters, temperature (60, 65, and 70°C) and time (12, 24, and 36 h) on the *L*a*b** colour profile, cooking loss, instrumental textural parameters were analysed by two-way analysis of variance (ANOVA) method using full factorial model (IBM SPSS Statistics for Windows, Version 26.0. Armonk, NY: IBM Corp, USA). The post hoc test for multiple comparisons were done by Student-Newman-Keuls (SNK) tests for temperature and time when there is no significant ($P > 0.05$) interaction. When there was a significant interaction ($p < 0.05$) multiple pair wise comparisons for significant temperature or time effects were analysed using LSD multiple comparison for individual time or temperature with adjusted level of significance of 1.6667%. The degree of meat and bone attachment was analysed by nonparametric Kruskal Wallis test and pair wise comparison were done at significance adjusted by Bonferroni correction.

The optimisation of sous vide temperature and time for response variable was carried out by fitting a third order multiple regression model (Eq. 2) using Minitab Version 18.1, Minitab, Inc., USA.

$$Y = \beta_0 + \beta_1X_1 + \beta_2X_2 + \beta_3X_1X_2 + \beta_4X_1^2 + \beta_5X_2^2 + \beta_6X_1^2X_2 + \beta_7X_1X_2^2 + \beta_8X_1^3 + \beta_9X_2^3 + e$$

2

Where, Y is the response variable, X_1 and X_2 are sous vide temperature and time, respectively; β_0 is the coefficient or constant terms. The linear coefficients for X_1 and X_2 are β_1 and β_2 , respectively; linear interaction coefficient is β_3 . β_4 and β_5 are quadratic coefficients; β_6 and β_7 are higher order interaction coefficients for X_1 and X_2 , respectively; β_8 and β_9 are cubic coefficients; e is the error term. The model was simplified by backward elimination, with an elimination criteria of $\alpha = 0.05$, to remove insignificant terms. The optimal area was determined from an overlaid contour plot of the significant response variable terms and optimum value of sous vide process variable temperature and time were calculated by response optimizer using simplified model for cooking loss, WBSF, TPA hardness, hue angle and Chroma and change in Chroma after and before sous vide.

3 Results And Discussion

3.1 Effect of different packaging materials on colour of cooked meat

The effect of different packaging materials on the colour of sous vide treated short ribs was investigated at the beginning of this study. The results showed that the use of triple layer Excel foil laminates could retain the meat colour during sous vide compared to polybag pouches (Fig. 1). When heating the meat in Excel foil laminates either at 60°C for 12, 24, or 36 h, or at 65°C for 12 h, the colour of cooked meat was light pink. After heating at 65°C for 24 and 36 h, the meat colour turned brown. The discolouration occurred due to oxidation of myoglobin to metmyoglobin (Abril et al., 2001; Roldán et al., 2013). Heating at 70°C caused the meat colour to progressively become browner with as the cooking time was increased. Heating the meat in vacuum packed polybags caused the meat colour to turn a blackish- brown at all cooking time-temperature combinations due to the oxidation of myoglobin to metmyoglobin. Increased oxidation of myoglobin took place due to higher oxygen permeability of polybags (50 cc/m²/day) compared to that of Excel foil laminate (≤ 0.1 cc/m²/day) at 23°C. Thus, the Excel foil laminate bags were chosen for further sous-vide treatments.

3.2 Effect of sous vide temperature and time on the quality properties of beef short ribs

The most important meat quality parameters are colour, flavour, juiciness, and tenderness (Listrat et al., 2016). Consumers judge meat freshness based on colour whereas tenderness influences their repurchasing decision (Bekhit et al., 2016; Suwandy et al., 2015b). Therefore, a processing technology, which can improve tenderness and flavour, while preserving colour and minimising loss of water, is of significant value (Kantono et al., 2019). The effect of sous vide processing parameters, temperature and time, on the meat quality parameters such as shrinkage in volume (%), cooking loss (%), CIE colour parameters, and instrumental texture parameters were assessed by full factorial two way ANOVA.

There was a significant ($p < 0.05$) effect of sous vide temperature and time on the decrease in volume (shrinkage) and cooking loss (Table 1). The interaction effect for both parameters were not significant ($p > 0.05$). The degree of shrinkage (overall mean) in the short ribs significantly ($p < 0.05$) increased as the sous vide temperature was increased from 60 to 70°C. Similarly, the percentage shrinkage (overall mean) of the meat significantly ($p < 0.05$) increased when the sous vide time was increased from 12 to 24 h; but no difference was observed between 24 and 36 h. When comparing the level of shrinkage after 12 h sous vide across the three temperatures, processing at 70°C had significantly higher ($p < 0.05$) shrinkage than at 60°C. After 24 h, sous vide processing at 70°C had significantly higher ($p < 0.05$) shrinkage than at 65 or 60°C. However, after 36 h, a significant difference in shrinkage was not observed for meat samples processed at 60, 65 or 70°C. At 60°C, shrinkage was higher after 24 and 36 h sous vide processing time compared to 12 h. However, at 65 or 70°C, no significant difference in % shrinkage between 12, 24 or 36 h was observed.

Table 1
Effect of sous vide temperature and time on physical properties of beef short ribs

Parameters	Temp (°C)	Time (h)			Overall mean
		12	24	36	
Shrinkage in volume (%)	60	12.45 ± 8.74 ^{abX}	16.08 ± 7.96 ^{abXY}	24.48 ± 8.22 ^Y	17.67 ± 9.52 ^a
	65	17.94 ± 13.07 ^{bc}	23.62 ± 7.28 ^{ab}	25.24 ± 3.30	22.27 ± 9.11 ^b
	70	22.86 ± 9.20 ^{cX}	33.27 ± 6.71 ^{bY}	29.41 ± 9.17 ^{XY}	28.51 ± 9.24 ^c
Overall Mean		17.75 ± 11.03 ^X	24.33 ± 10.07 ^Y	26.38 ± 7.83 ^Y	
Cooking loss (%)	60	12.78 ± 2.15 ^{aX}	15.87 ± 2.53 ^{aY}	18.08 ± 1.47 ^{aY}	15.58 ± 3.00 ^a
	65	15.51 ± 3.07 ^{abX}	21.08 ± 1.52 ^{bY}	21.33 ± 2.61 ^{bY}	19.30 ± 3.63 ^b
	70	18.23 ± 4.25 ^{bX}	23.15 ± 3.58 ^{bY}	24.78 ± 2.30 ^{cY}	22.05 ± 4.39 ^c
Overall Mean		15.51 ± 3.90 ^X	20.03 ± 4.05 ^Y	21.39 ± 3.49 ^Y	
Soluble collagen (%)	60	20.19 ± 4.50 ^b	20.00 ± 3.33 ^b	19.31 ± 2.94 ^c	19.83 ± 3.48
	65	23.36 ± 3.67 ^{ab}	21.16 ± 5.39 ^b	27.05 ± 4.16 ^b	23.85 ± 4.91
	70	28.24 ± 8.27 ^{aY}	35.29 ± 6.39 ^{aX}	40.24 ± 4.39 ^{aX}	34.59 ± 8.00
Overall mean		23.93 ± 6.49	25.48 ± 8.66	28.86 ± 9.59	
Myofibrillar fragmentation index (MFI)	60	109.15 ± 14.46 ^{bY}	117.47 ± 14.65 ^{cXY}	129.29 ± 10.07 ^{bX}	118.85 ± 15.44 ^{cY}
	65	129.42 ± 18.51 ^a	138.54 ± 15.47 ^b	139.53 ± 15.90 ^b	135.83 ± 16.75 ^{bY}
	70	139.86 ± 15.05 ^{aY}	163.56 ± 14.96 ^{aX}	176.43 ± 12.53 ^{aX}	159.95 ± 20.63 ^{aX}
Overall mean		126.14 ± 20.24 ^Z	139.85 ± 24.03 ^Y	148.63 ± 23.96 ^X	
Values presented are means ± SD (n = 10 for cooking loss, shrinkage in volume and MFI, n = 7 for collagen solubilized)					
a-c values with different letters within in the same column differ significantly (p < 0.05)					
X-Z values with different letters within the same row differ significantly (p < 0.05)					
Values without the superscripts do not differ significantly					

There was a significant ($p < 0.05$) increase in cooking loss (%) as sous vide temperature and time were increased. The overall mean cooking loss (%) of short ribs sous vide processed at 70°C was significantly higher ($p < 0.05$) than ribs heated at 65 or 60°C; and at 65°C cooking loss was significantly higher than at 60°C. This temperature effect was also observed in meat processed for 12 h. However, after 24 h SV the only significant cooking loss effect was observed between meats processed at 70°C and 65°C with meat processed at 60°C and after 36 h no temperature effect on cooking loss was observed. Meat processed at 24 and 36 h had a significantly higher cooking loss than meat sous vide processed for 12 h. The cooking losses found in this study were lower compared to other studies on tough meat cuts, even at 70°C (Bhat et al., 2020; Christensen et al., 2013; Ismail et al., 2019; Rinaldi et al., 2014). Bhat et al. (2020) reported cooking loss (%) of 28.67 ± 0.61 in beef semitendinosus muscle of dairy cows. Christensen et al. (2013) reported 26.6 and 25.9% cooking loss at 60°C for 10 h for cows (4–6 years old) and young bulls (10–12 months), respectively. Ismail et al. (2019) reported 29.07% cooking loss at 60°C for 12h for semitendinosus

muscles of Hanwoo steers (24–32 months). Rinaldi et al. (2014) reported 38.9% cooking loss at 75°C for 36 h for beef semitendinosus muscle. However, the cooking losses in this study may have been reduced due to limited cooking loss of the bone present in the short rib sections.

The shrinkage in volume reduced the water holding capacity (WHC) of the meat and increased the cooking loss, which is associated with a decrease in juiciness and succulence (Arroyo, Eslami, et al., 2015; Dominguez-Hernandez et al., 2018). The WHC of meat also affects its texture, tenderness, and flavour (Arroyo, Eslami, et al., 2015; Roldán et al., 2013). The shrinkage in volume and cooking loss were positively correlated (Pearson correlation coefficient 0.546, $p < 0.01$). Both significantly increased when sous vide temperature and cooking time were increased. The likely mechanism for the higher cooking loss at higher sous vide temperature and longer cooking time was the increased myofibrillar shrinkage and contraction. This phenomenon decreased the intracellular space and volume between strands of actin and myosin of sarcomere, and this consequently increased the expulsion of intracellular water (Baldwin, 2012; Bertram et al., 2006; Bertram et al., 2002; Cheng & Sun, 2008; Micklander et al., 2005; Suwandy et al., 2015a). Intracellular water accounts about 70–75% of total water content of meat (Bertram et al., 2006). Furthermore, the higher the cooking temperature will be the greater the denaturation of myofibrillar proteins will be. As myofibril proteins hold the major proportion of water in meat, their denaturation, especially of myosin, reduces the meat's water holding capacity (Wiklund et al., 2001). Bertram et al. (2006) also showed a significant relationship between the increase in denaturation of myofibrillar proteins, seen by major changes in the FT-IR spectroscopic bands in the amide I-region ($1700 - 1600 \text{ cm}^{-1}$) and broadening of T_{21} (35–50 ms) NMR relaxometry peak, and a decrease in myofibrillar water during heating.

The effect of sous vide temperature ($p < 0.001$) and time ($p = 0.008$) on the proportion of soluble collagen relative to total collagen in the ribs were significant as was the interaction between sous vide temperature and time ($p = 0.01$) (Table 1). Therefore, the effect of temperature on soluble collagen of short ribs was affected by the level of processing time and vice versa. The soluble collagen of ribs cooked at 70°C had significantly higher than when cooked at 65 or 60°C; and in addition, short ribs cooked at 65°C were significantly higher than when cooked at 60°C. This effect was observed for ribs sous vide for 36 h but in ribs cooked for 24 h soluble collagen was only significantly higher at 70°C than the other temperatures, and at 12 h soluble collagen was significantly lower at 60°C than at 65°C or 70°C. Similarly, the effect of sous vide temperature ($p < 0.001$) and time ($p < 0.001$) on the MFI were significant but the interaction term was not significant ($p > 0.05$) (Table 1). The MFI values increased with an increase in sous vide temperature and time.

The tenderness of meat and meat products has a dramatic impact on consumer's repurchasing decisions (Bolumar et al., 2013). The tenderness of a meat cut depends on the amount of connective tissue present, the extent of proteolysis of the myofibrillar proteins, as well as animal (age, breed), environmental, and genetic factors (Anderson et al., 2012). The effect of sous vide temperature and time on tenderness and texture parameters of short ribs instrumental parameters were assessed using WBSF and TPA parameters such as hardness, adhesiveness, cohesiveness, springiness, gumminess, chewiness, and resilience (Table 2). There was a significant effect of temperature ($p < 0.05$) on WBSF, hardness, cohesiveness, gumminess and chewiness; and a significant effect of time ($p < 0.05$) on WBSF, hardness, adhesiveness, cohesiveness, springiness, gumminess chewiness and resilience of sous vide ribs.

WBSF (N), which is correlated to the shearing and compression during eating meat, was significantly affected by temperature and time ($p = 0.026$ and $p < 0.001$, respectively) (Table 2). However, a significant time – temperature interaction was observed ($p = 0.010$) indicating that the WBSF did not change the same way with temperature as time was changed. At sous vide time of 12 h, only ribs processed at 60°C had significantly ($p < 0.05$) lower WBSF value than at 65 and 70°C and after 24 h sous vide processing, no significant difference between the three temperatures were observed. However, at 36 h sous vide, a significant difference in WBSF was observed between ribs processed at 60 and 70°C. However, WBSF values at all temperature and time combinations were lower than the threshold level for beef to be described as tender. Destefanis et al. (2008) and Dufey et al. (2017) concluded separately from consumer testing that beef meat with WBSF $< 42.87 \text{ N}$ and $< 32.57 \text{ N}$ ($\leq 3.3 \text{ kg force}$), respectively can be termed tender.

The trends observed for TPA hardness values of sous vide ribs were similar to the WBSF values. In TPA, hardness is maximum force encountered by the first depression of the probe, which is analogous to the hardness encountered upon the first bite; however hardness perception continues during the subsequent mastication process (Nishinari & Fang, 2018). As with WBSF a significant temperature-time interaction ($p = 0.022$) was observed for hardness values. The hardness was significantly lower after sous vide of ribs for 24 and 36 h and at 70°C compared to sous vide for 12 h, at 60°C or 65°C (Table 2). No difference was observed in hardness between ribs sous vide for 12 h at the three sous vide temperatures. However, after sous vide for 24 or 36 h the hardness of meat sous vide at 70°C was significantly lower than for sous vide at 60°C.

Significantly lower WBSF values for sous vide treatment at 60°C than at 65 or 70°C after 12 h may be associated with lower degree of myofibrillar protein denaturation and lower level of shrinkage and cooking loss. It is well documented that as the cooking temperature increases the degree of myofibrillar proteins denaturation increases, as does the shrinkage of the sarcomere reducing its volume thereby resulting in meat becoming tough and dry. Higher cooking losses resulted in the tough meat (Roldán et al., 2013). The lower WBSF and hardness values in sous vide ribs after processing for 36h at 70°C compared to 60°C might be due to the higher level of solubilisation and gelation of collagen and sarcoplasmic proteins even though the extent of denaturation may be similar.

Table 2
Effect of sous vide temperature and time on instrumental textural parameters of beef short ribs

Parameters	Temp (°C)	Time (Hours)			Overall mean
		12	24	36	
Warner Bratzler shear force (WBSF)	60	21.02 ± 5.64 ^{bX}	19.35 ± 4.24 ^{XY}	17.59 ± 4.46 ^{aY}	19.32 ± 4.97
	65	24.68 ± 5.99 ^{aX}	21.09 ± 5.60 ^Y	17.08 ± 4.51 ^{abZ}	20.95 ± 6.19
	70	24.41 ± 5.79 ^{aX}	18.29 ± 5.02 ^Y	14.23 ± 5.22 ^{bZ}	18.98 ± 6.76
Overall mean		23.37 ± 5.98	19.57 ± 5.06	16.30 ± 4.92	
TPA Hardness (N)	60	22.94 ± 11.44	21.96 ± 6.93 ^a	20.81 ± 7.53 ^a	21.90 ± 8.85
	65	21.12 ± 10.64	19.66 ± 5.88 ^{ab}	22.05 ± 6.91 ^a	20.94 ± 8.08
	70	21.51 ± 8.64 ^X	16.90 ± 6.25 ^{bY}	15.26 ± 4.03 ^{bY}	17.89 ± 7.06
Overall mean		21.86 ± 10.27	19.51 ± 6.66	19.37 ± 6.96	
TPA Adhesiveness (N.sec)	60	-0.02 ± 0.03	-0.02 ± 0.04	-0.03 ± 0.05	-0.03 ± 0.04
	65	-0.02 ± 0.06	-0.02 ± 0.04	-0.03 ± 0.05	-0.03 ± 0.05
	70	-0.03 ± 0.05 ^Y	-0.03 ± 0.05 ^Y	-0.06 ± 0.09 ^X	-0.04 ± 0.07
Overall mean		-0.03 ± 0.05 ^Y	-0.02 ± 0.05 ^Y	-0.04 ± 0.07 ^X	
TPA Cohesiveness	60	0.45 ± 0.07 ^{aX}	0.43 ± 0.05 ^{aX}	0.38 ± 0.08 ^{aY}	0.42 ± 0.07
	65	0.43 ± 0.09 ^{abX}	0.34 ± 0.08 ^{bY}	0.36 ± 0.04 ^{aY}	0.38 ± 0.08
	70	0.40 ± 0.08 ^{bX}	0.32 ± 0.06 ^{bY}	0.29 ± 0.05 ^{bZ}	0.33 ± 0.08
Overall mean		0.42 ± 0.08	0.36 ± 0.08	0.34 ± 0.07	
TPA Springiness	60	0.61 ± 0.09	0.59 ± 0.08	0.58 ± 0.11	0.59 ± 0.09
	65	0.63 ± 0.10	0.62 ± 0.09	0.59 ± 0.10	0.61 ± 0.10
	70	0.68 ± 0.11 ^X	0.60 ± 0.13 ^Y	0.58 ± 0.09 ^Y	0.62 ± 0.12
Overall mean		0.64 ± 0.10	0.60 ± 0.10	0.58 ± 0.10	0.61 ± 0.10
TPA Gumminess	60	10.39 ± 5.65 ^X	9.67 ± 3.79 ^{aXY}	8.33 ± 3.99 ^{aY}	9.46 ± 4.60
	65	9.47 ± 5.88 ^X	6.93 ± 3.02 ^{bY}	8.06 ± 2.92 ^{aXY}	8.15 ± 4.28
	70	8.73 ± 4.67 ^X	5.63 ± 2.63 ^{bY}	4.42 ± 1.43 ^{bY}	6.26 ± 3.67
Overall mean		9.53 ± 5.43	7.41 ± 3.58	6.94 ± 3.45	
TPA Chewiness (N)	60	6.41 ± 3.67 ^X	5.72 ± 2.44 ^{aXY}	5.02 ± 2.81 ^{aY}	5.72 ± 3.05
	65	6.14 ± 4.13 ^X	4.28 ± 1.89 ^{bY}	4.84 ± 2.04 ^{aXY}	5.09 ± 2.96
	70	5.97 ± 3.36 ^X	3.48 ± 1.77 ^{bY}	2.51 ± 0.81 ^{bY}	3.99 ± 2.66
Overall mean		6.18 ± 3.71	4.49 ± 2.24	4.13 ± 2.35	
TPA Resilience	60	0.27 ± 0.05 ^{aX}	0.22 ± 0.04 ^{aY}	0.19 ± 0.04 ^{aZ}	0.23 ± 0.05
	65	0.23 ± 0.06 ^{bX}	0.17 ± 0.04 ^{bY}	0.17 ± 0.03 ^{aY}	0.19 ± 0.05
	70	0.22 ± 0.05 ^{bX}	0.15 ± 0.03 ^{cY}	0.13 ± 0.02 ^{bZ}	0.16 ± 0.05
Overall mean		0.24 ± 0.06	0.18 ± 0.05	0.16 ± 0.04	0.19 ± 0.06

Values presented are means ± SD (n = 30 for WBSF and n = 50 TPA parameters, with 10 short ribs with 3 and 5 samples per meat pieces for WBSF and TPA, respectively)

^{a-c} values with different letter within in the same column differ significantly (p < 0.05)

^{X-Z} values with different uppercase letters within the same row differ significantly (p < 0.05)

Values without the superscripts do not differ significantly

Similar results were observed by Vaudagna et al. (2002) and García-Segovia et al. (2007). Vaudagna et al. (2002) reported lower WBSF at 65°C than at 50°C. García-Segovia et al. (2007) reported a significant decrease in WBSF with an increase in sous vide temperature from 60 to 80°C. This was credited to moist in-pack environment and overpressure created by saturated steam resulting in improved heat transfer, which led to greater unfolding and gelation of sarcoplasmic protein and myofibrillar protein as well as increased breakage of thermolabile bonds of triple helix structure of collagen, which increased its solubilisation (García-Segovia et al., 2007; Tornberg, 2005). This is supported by the observation that the percentage of soluble collagen was significantly higher ($p < 0.05$) at 70°C than at 60°C after 36 h sous vide processing (Table 1). In addition, Zielbauer et al. (2016) reported that the denaturation temperature and enthalpy is lower than the dry heating denaturation temperature and enthalpy of myofibrillar proteins during moist heating techniques, like sous vide emphasising that the same level of denaturation could be achieved at lower temperatures. The importance of solubilisation of collagen for enhancement of tenderness can be stressed from the observation by Alahakoon et al. (2018). They did not observe the significant increase in the amount of soluble collagen of beef brisket when sous vide temperature was increased from 60°C to 70°C processed for same time of 24 h and no subsequent decrease in WBSF values at that sous vide condition.

Meat tenderness is not only affected by the distribution and amount of connective tissues but also affected by the overlapping and disintegration of the myofibrillar structure (Aroeira et al., 2020; Sawdy et al., 2004). It is also stated that increase in tenderness is also due to higher degree of meat proteolysis indicated by MFI (Aroeira et al., 2020; Culler et al., 1978). The results discussed in the current study also agree with the above argument. Significantly lower WBSF and TPA hardness and significantly higher MFI was observed with increase in sous vide temperature and time.

For adhesiveness, which is the force required to separate the plunger and meat (Nishinari & Fang, 2018) indicating meat stickiness due to increased mass transfer towards the surface, short ribs sous vide for 36 h had significantly lower values than that SV for 12 or 24 h. This difference in overall mean was due to the difference in adhesiveness at sous vide time of 36h at 70°C which was significantly lower than 12 and 24h sous vide at 70°C.

TPA cohesiveness, which relates to how much the meat regains its structure and is associated with the integrity of meat was found to have significant time ($p < 0.001$), temperature ($p < 0.00$) and time- temperature interaction ($p = 0.035$). Regardless of whether meat was sous vide at 60, 65 or 70°C, cohesiveness was significantly lower at 36 h than after 12h. When assessing the effect of temperature at the same processing time, sous vide at 70°C the cohesiveness is significantly lower than at 60°C for 12, 24 or 36 h. These results indicate that the textural coherence and integrity decrease at SV temperature greater than or equal to 65°C and at SV time of 36 h.

Springiness, previously called elasticity, of sous vide ribs, which is the rate at which deformed ribs return to its undeformed condition after the deforming force is removed, displayed a significant sous vide time-temperature interaction ($p = 0.035$) in addition to the significant ($p < 0.001$) time effect. Short ribs processed for 24 or 36 h at 70°C had significantly lower springiness than that cooked for 12 h. Similarly, the sous vide time-temperature interaction was also significant ($p < 0.05$) for TPA gumminess. Short ribs sous vide processed for 24 or 36 h had significantly gumminess lower than 12 h and these differences were also observed at 60, 65 or 70°C. Gumminess of ribs sous vide at 70°C was significantly lower than at 65 or 60°C after sous vide for 12 h. This difference was observed in ribs sous vide for 24 or 36 h. Gumminess, energy required to disintegrate a semisolid food product to a state ready for swallowing (Nishinari & Fang, 2018), is derived from TPA hardness and cohesiveness. Therefore, based on the findings for hardness and cohesiveness, springiness and gumminess will decrease as the sous vide temperature is increased and sous vide time lengthened.

TPA chewiness is calculated from the primary parameters of hardness, cohesiveness, and elasticity (Nishinari & Fang, 2018). As the temperature was increased from 60°C to 70°C chewiness significantly decreased and similarly increasing the sous vide time from 12 h to 24 or 36 h significantly reduced the chewiness. At longer sous vide times of 24 or 36 h, higher sous vide temperature of 70°C had significantly lower chewiness than at 60°C. This effect was not observed at a sous vide time of 12h, potentially reflecting the significant SV time and temperature interaction ($p = 0.011$). The same trends were observed for the resilience values where they significantly decreased in ribs sous vide for longer times at higher temperatures

Meat colour is a critical quality attribute as the visual appearance of raw meat influences the consumer's decision to purchase and the acceptance of the cooked meat (Arroyo, Lascorz, et al., 2015). The colour of fresh meat depends on the content and physicochemical state of myoglobin, i.e. purple (reduced myoglobin), red (oxymyoglobin), and brown (metmyoglobin) (Abril et al., 2001). The effect of sous vide temperature and time on CIE L^* , a^* , b^* , Chroma ($C^* = \sqrt{a^{*2} + b^{*2}}$), and hue angle ($h = \tan^{-1} \frac{b^*}{a^*}$) are summarised in Table 3. Significant ($p < 0.05$) sous vide time and temperature effects were observed for L^* , significant effect of time were observed for h , and significant ($p < 0.05$) temperature effects for b^* , C^* , Δb^* and ΔE^* . An interaction of sous vide time and temperature on the colour of ribs was observed for C^* . These were the only colour parameters for which significant effects were observed. L^* values of the sous vide rib surface were significantly higher at 60°C than at 65 or 70°C and ribs sous vide at 65°C higher than at 70°C. Similarly, 12 h sous vide had significantly higher L^* values than for 24 or 36 h sous vide. The increase in the change in lightness (ΔL^*), which is a comparison between the meat before and after sous vide were significantly higher at 60°C than at 65 or 70°C. The higher lightness associated with ribs sous vide at lower temperature and shorter times is due to higher amount of free water on the surface (Sánchez del Pulgar et al., 2012), which is consistent with the cooking loss results.

Table 3
Effect of sous vide temperature and time on CIE L*a*b*, h and C* colour parameters of beef short ribs

Parameters	Temp (°C)	Time (Hours)			Overall mean
		12	24	36	
Lightness (L*) after cooking	60	53.41 ± 1.76 ^{aX}	51.27 ± 4.09 ^{aX}	49.79 ± 4.25 ^{aY}	51.49 ± 3.75 ^a
	65	51.53 ± 4.27 ^{aX}	45.56 ± 3.98 ^{bY}	46.72 ± 3.12 ^{abY}	47.94 ± 4.53 ^b
	70	47.59 ± 2.46 ^{bX}	43.83 ± 3.90 ^{bX}	43.68 ± 3.73 ^{bX}	45.03 ± 3.79 ^c
Overall mean		50.84 ± 3.82 ^X	46.88 ± 5.03 ^Y	46.73 ± 4.40 ^Y	
Redness (a*) after cooking	60	12.01 ± 2.87	10.68 ± 1.26	11.77 ± 1.61	11.49 ± 2.05
	65	9.13 ± 2.35	11.77 ± 1.77	11.44 ± 1.83	10.78 ± 2.27
	70	11.16 ± 1.34	11.06 ± 1.46	11.47 ± 1.42	11.23 ± 1.37
Overall mean		10.77 ± 2.52	11.17 ± 1.53	11.56 ± 1.58	
Yellowness (b*) after cooking	60	17.28 ± 2.58 ^a	15.38 ± 0.86	15.53 ± 1.00	16.06 ± 1.84 ^a
	65	14.31 ± 2.66 ^b	14.89 ± 0.99	14.94 ± 1.08	14.71 ± 1.72 ^b
	70	15.07 ± 0.65 ^b	14.52 ± 0.85	14.47 ± 1.04	14.69 ± 0.88 ^b
Overall mean		15.55 ± 2.46	14.93 ± 0.94	14.98 ± 1.10	
Hue angle after cooking	60	55.65 ± 3.77	55.31 ± 2.66	52.98 ± 2.40	54.65 ± 3.14
	65	57.70 ± 3.86 ^X	51.82 ± 4.54 ^Y	52.76 ± 3.66 ^Y	54.10 ± 4.69
	70	53.58 ± 3.42	52.82 ± 4.21	51.68 ± 4.10	52.69 ± 3.87
Overall mean		55.64 ± 3.95 ^X	53.32 ± 4.04 ^Y	52.48 ± 3.39 ^Y	
Chroma after cooking	60	21.08 ± 3.65 ^a	18.74 ± 1.24	19.50 ± 1.71	19.77 ± 2.55
	65	17.01 ± 3.38 ^b	19.03 ± 1.38	18.85 ± 1.76	18.30 ± 2.44
	70	18.78 ± 0.98 ^b	18.30 ± 1.02	18.51 ± 1.18	18.53 ± 1.05
Overall mean		18.95 ± 3.29	18.69 ± 1.22	18.95 ± 1.58	
Change in L*	60	11.02 ± 6.63 ^a	11.34 ± 6.41 ^a	11.49 ± 6.86 ^a	11.28 ± 6.41 ^a
	65	7.94 ± 5.94 ^a	7.14 ± 6.88 ^{ab}	6.85 ± 6.64 ^{ab}	7.31 ± 6.29 ^b
	70	4.35 ± 5.67 ^a	4.28 ± 4.63 ^b	3.97 ± 7.78 ^b	4.20 ± 5.95 ^b
Overall mean		7.77 ± 6.50	7.58 ± 6.54	7.44 ± 7.55	
Change in a*	60	-3.12 ± 1.90	-5.763 ± 2.75	-2.98 ± 2.81	-3.96 ± 2.76
	65	-5.54 ± 3.88	-4.27 ± 2.17	-3.55 ± 2.67	-4.45 ± 3.01
	70	-4.28 ± 1.13	-4.84 ± 2.06	-4.06 ± 2.78	-4.39 ± 2.05
Overall mean		-4.31 ± 2.87	-4.96 ± 2.35	-3.53 ± 2.69	
Change in b*	60	4.64 ± 3.02 ^a	2.13 ± 2.21	3.44 ± 1.96	3.40 ± 2.57
	65	1.25 ± 3.58 ^b	1.47 ± 2.91	2.79 ± 2.51	1.83 ± 3.00
	70	0.90 ± 2.71 ^b	1.53 ± 1.58	2.41 ± 3.05	1.61 ± 2.52
Overall mean		2.26 ± 3.47	1.71 ± 2.24	2.88 ± 2.49	

Values presented are means ± SD (n = 10 for each treatment)

The change in L*, a*, b*, hue angle, and chroma refer to the difference in value after and before sous vide

^{a-c} values with different letter within in the same column differ significantly (p < 0.05)

^{X-Z} values with different letters within the same row differ significantly (p < 0.05)

Values without the superscripts do not differ significantly (p > 0.05)

Parameters	Temp (°C)	Time (Hours)			Overall mean
		12	24	36	
Change in hue angle	60	15.89 ± 7.19	16.28 ± 5.86	13.73 ± 7.03	15.30 ± 6.58
	65	15.98 ± 6.95	12.23 ± 4.39	13.51 ± 7.26	13.91 ± 6.31
	70	11.31 ± 6.44	13.53 ± 4.17	13.75 ± 8.51	12.86 ± 6.48
Overall mean		14.39 ± 6.99	14.02 ± 5.00	13.66 ± 7.36	
ΔE^*	60	12.80 ± 6.65 ^a	13.72 ± 5.41 ^a	12.81 ± 6.81	13.11 ± 6.11 ^a
	65	11.35 ± 5.11 ^a	10.33 ± 4.62 ^{ab}	9.66 ± 5.36	10.45 ± 4.91 ^b
	70	7.66 ± 4.22 ^a	7.93 ± 2.70 ^b	9.54 ± 4.32	8.37 ± 3.78 ^b
Overall mean		10.60 ± 5.67	10.66 ± 4.88	10.67 ± 5.61	
ΔH^*	60	5.49 ± 2.85	5.69 ± 2.27	4.54 ± 2.44	5.24 ± 2.49
	65	4.99 ± 2.11	4.24 ± 1.52	4.51 ± 2.53	4.58 ± 2.04
	70	3.83 ± 2.07	4.56 ± 1.40	4.64 ± 3.00	4.34 ± 2.21
Overall mean		4.77 ± 2.39	4.83 ± 1.82	4.56 ± 2.57	
Values presented are means ± SD (n = 10 for each treatment)					
The change in L*, a*, b*, hue angle, and chroma refer to the difference in value after and before sous vide					
a-c values with different letter within in the same column differ significantly (p < 0.05)					
X-Z values with different letters within the same row differ significantly (p < 0.05)					
Values without the superscripts do not differ significantly (p > 0.05)					

The overall mean value of hue angle of ribs sous vide for 12h was significantly higher than those sous vide for 24 or 36 h at 65°C. C* of ribs sous vide at 60 °C was significantly higher of ribs sous vide at 65 or 70°C. As C* and hue angle values explain the colour intensity and the relative position of colour between redness and yellowness, respectively (Bekhit et al., 2001) they are good indicators of metmyoglobin formation and increase in meat brownness (Khan et al., 2017; Suwandy et al., 2015c). The hue angle value generally increases because of myoglobin and oxymyoglobin oxidation (Bekhit & Faustman, 2005). The extent of oxidation of myoglobin depends on temperature, oxygen pressure, and lipid oxidation. The rate of myoglobin oxidation increases when the temperature is increased (Faustman et al., 2010). Examining the overall change in colour (ΔE^*) ribs sous vide at 60°C sous vide had a significantly higher ΔE^* value than at 65 or 70°C.

The attachment of the meat and bone after sous vide was measured on an ordinal scale from 1–5 (strongest to weakest attachment, respectively). The meat of ribs sous vide at 70°C for 36 h had become more detached from the bone than all other sous vide conditions, except ribs sous vide at 65°C for 36 h, based on a significantly higher median and mean rank (Table 4). At all other SV conditions no significant reduction in the attachment of meat to bone occurred. Having said this, all other sous vide time and temperature combinations, except ribs sous vide at 60°C for 12, 24 h, and ribs sous vide at 65°C for 12 h, contained at least one sample where the meat was attached less tightly to the bone.

Table 4
Loosening of meat and bone at different sous vide temperature and time

Temp(°C)-Time (h)	Mean Rank	Median	Range
60 - 12	31.50	1.00 ^a	1-1
60 - 24	31.50	1.00 ^a	1-1
60 - 36	49.30	1.00 ^a	1-3
65 - 12	31.50	1.00 ^a	1-1
65 - 24	42.15	1.00 ^a	1-2
65 - 36	60.55	2.00 ^{ab}	1-4
70 - 12	35.05	1.00 ^a	1-2
70 - 24	48.10	1.00 ^a	1-4
70 - 36	79.85	5.00 ^b	1-5

Note: The loosening of meat and bone are on the ordinal scale of 1-5, Where:
 1: Meat and bone completely intact
 5: Meat and bone completely separated
 n = 10; Kruskal-Wallis H value 46.667, $p < 0.001$
^{a-b} values with different letter within in the same column differ significantly ($p < 0.05$)

3.3 Optimization of sous vide temperature and time for short ribs

Optimization of sous vide temperature and time was carried out by fitting a third order multiple regression model using cooking loss (%), WBSF, TPA parameters, hue angle, and Chroma as response variables. The model was significant ($p < 0.05$) for all responses, except adhesiveness (Table 5). The lack of fit, which measures the ability of the model to describe the variability of data, was not significant ($p = 0.3-0.8$) reiterating that the regression equations adequately describe the data and predict the responses at any combination of sous vide temperature and time without significant deviation from the expected values.

The association of cooking loss (%) with the significant positive linear terms for time and temperature reflect that cooking loss increases as sous vide temperature and time increase. The significant quadratic term for time reflects that the rate of increase in cooking loss decreases as sous vide time increases (Fig. 2A). The significant negative coefficient of quadratic term of temperature and negative time - temperature interaction term for WBSF shows that the WBSF potentially reaches maximum that varies between 65 to 70°C depending upon sous vide time (Fig. 2B). In addition the rate of decrease in WBSF with time increases as the temperature increases.

Table 5
Second order regression model and coefficients of sous vide temperature and time for various response variable

Terms	Cooking loss (%)	WBSF (N)	Hardness (N)	Adhesiveness (N.sec)	Cohesiveness	Springiness	Gumminess	Chewiness	Resilience	Lightness (L*)
Constant (β)	-34.28	-353	952	-0.038	1.038	0.6809	33.55	19.19	0.768	402
Linear										
T(°C)	0.648**	11.04	-29.1*		-0.009**		-0.347**	-0.1826**	-0.007**	-10.30***
t(h)	0.772**	1.517*	-41.70	0.001	-0.003**	0.0028**	-0.127**	-0.0997	-0.01**	0.269***
Quadratic										
T(°C)* T (°C)		-0.080*	0.227							0.076*
t (h)*t (h)	-0.011*								0.0001**	
Interaction terms										
T(°C)*t (h)		-0.028*	1.314*							
T(°C)*T(°C)*t(h)			-0.0134*							
T(°C)*t(h)*t (h)										
Regression p Value	< 0.001	< 0.001	0.003	0.074	< 0.001	0.001	< 0.001	< 0.001	< 0.001	< 0.001
Lack of fit	0.651	0.803	0.421	0.859	0.256	0.206	0.31	0.141	0.538	0.676
R ² (adjusted)	64.13	34.50	13.99	2.48	41.58	10.94	26.04	24.42	65.12	44.66
R ² (predicted)	61.97	29.72	6.48	0	38.80	7.62	22.24	20.52	63.10	41.52

Note: T(°C): Sous vide temperature ((°C); t (h): Sous vide time (hours)

* indicates the term is significant at $p < 0.05$; ** indicates the term is significant at $p < 0.01$, ***indicates the term is significant at $p < 0.001$

The linear and quadratic interaction terms in addition to the negative coefficient time and temperature terms for hardness indicates that the decrease in hardness with increase in sous vide time and temperature is complex (Fig. 2C). As sous vide temperature increases the change in hardness of the ribs with increasing time goes from a very slight decrease at 60°C, to a slight increase at 65°C, to a large decrease at 70°C. In addition, at 12 h sous vide hardness of the ribs appears to be at a minimum at 65°C. Significant negative coefficients for temperature and time observed for cohesiveness and gumminess, showed that these texture parameters decreased with increasing temperature and time. For resilience in addition to the negative coefficients for sous vide temperature and time there was a significant quadratic term for sous vide time. This suggests that as the sous vide time and temperature increased the resilience decreased and the rate of decrease with time slowed at time increased. The only significant term for chewiness was negative linear term for temperature indicating that as sous vide temperature increased chewiness of the ribs decreased and that sous vide time did not have a significant effect on chewiness of the ribs.

The significant negative sous vide temperature, time and quadratic temperature terms for the instrumental colour parameter, L* indicate that as temperature and time increased L* decreased though the rate of decrease in L* became smaller as the temperature approached to 70°C (Fig. 2F).

Similarly, the significant and negative linear coefficient of SV time and temperature showed that hue decreased with increasing temperature and time (Fig. 2G). The positive and significant quadratic terms of C* for time explained that the minima reached at around 24 h and further increase in C* after that time (Fig. 2H). The change in chroma after and before sous vide (ΔC^*) had significant negative term for linear term for time and positive quadratic term for temperature, which depicted that the ΔC^* is lowest at 65°C and time above 34 h. Significant positive linear interaction between temperature and time and quadratic terms of temperature for soluble collagen (%) showed that the rate of its increase enhanced with elevating in temperature and prolonging sous vide time (Fig. 2I).. However, the change in hue angle after and before sous vide (Δh) had no significant terms for temperature and time.

The optimal sous vide temperature and time to achieve the highest tenderness, and lowest hue angle, C*, and ΔC^* while achieving the lowest cooking loss was calculated by response optimizer to be 60°C and 34.06 h, respectively with an optimal desirability of 0.53. This resulted in cooking loss (%), WBSF, TPA hardness, hue angle, C*, and ΔC^* of 18.15%, 17.65 N, 20.79 N, 54.58, 18.15, and - 1.98, respectively. These values are very near to the experimental values of cooking loss (%), WBSF, TPA hardness, hue angle, and C* for sous vide of ribs at 60°C for 36 h, which were 17.71%, 18.51N, 21.091N, 55.01, 17.95, respectively. Under the optimum sous vide conditions, the attachment of the meat to the bone was not significantly different from the lowest rank value. This is supported by an overlay of all the contour plots which shows the wider optimal window for sous vide conditions of ribs to be a time of 24 to 35 h and temperature of 60 to 63°C (Fig. 3).

4 Conclusion

The shrinkage in volume (%), cooking loss (%), soluble collagen (%) and MFI increased significantly ($p < 0.05$) with an increase in temperature or time. There was a significantly lower WBSF and TPA hardness value at 70°C than at 60°C for 36 h sous vide processing, due to an increase in the level of solubilisation

and gelation of collagen and sarcoplasmic proteins. This might be credited to moist in-pack environment improved heat transfer due to overpressure created by saturated steam. The effect of sous vide temperature and time was not significant ($p > 0.05$) on CIE hue angle (h) and increase in hue angle (Δh) may have been due to the lower oxygen permeability of sous vide laminate pouch which reduced the oxidation of red oxymyoglobin and deoxymyoglobin to brown metmyoglobin. Based on a third order multiple regression model, the optimum sous vide processing regime was found at 60°C for 34.06 h resulting in the highest tenderness and colour parameters whilst minimizing cooking loss and achieving a cooked rib where meat and bone were well attached. These conditions are recommended for commercial sous vide of beef short ribs.

Declarations

The authors acknowledge funding from the New Zealand Ministry of Business, Innovation and Employment (contract MAUX1402) for the Food Industry Enabling Technologies (FIET) programme. There are no competing interests that are directly or indirectly related to the work submitted for publication

Acknowledgement and Funding

This work was supported by the Food Industry Enabling Technologies (FIET) program funded by the New Zealand Ministry of Business, Innovation and Employment (contract MAUX1402). Oey is affiliated to the Riddet Institute, a New Zealand Centre of Research Excellence, funded by the Tertiary Education Commission.

Conflict of interest

The authors declare no conflict of interest.

Authors Contribution

Roman Karki: Data curation, Methodology, Software, Formal analysis, Investigation, Validation, Visualization, Roles/Writing - original draft, Conceptualization, Roles/Writing - original draft and editing. **Phil Bremer:** Conceptualization, Funding acquisition, Investigation, Methodology, Project administration, Writing - review and editing. **Patrick Silcock:** Conceptualization, Funding acquisition, Investigation, Methodology, Project administration, Writing - review and editing. **Indrawati Oey:** Conceptualization, Funding acquisition, Investigation, Methodology, Project administration, Resources, Software, Supervision, Writing - review and editing.

Data availability

The data of the experiment are available upon request

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Figures

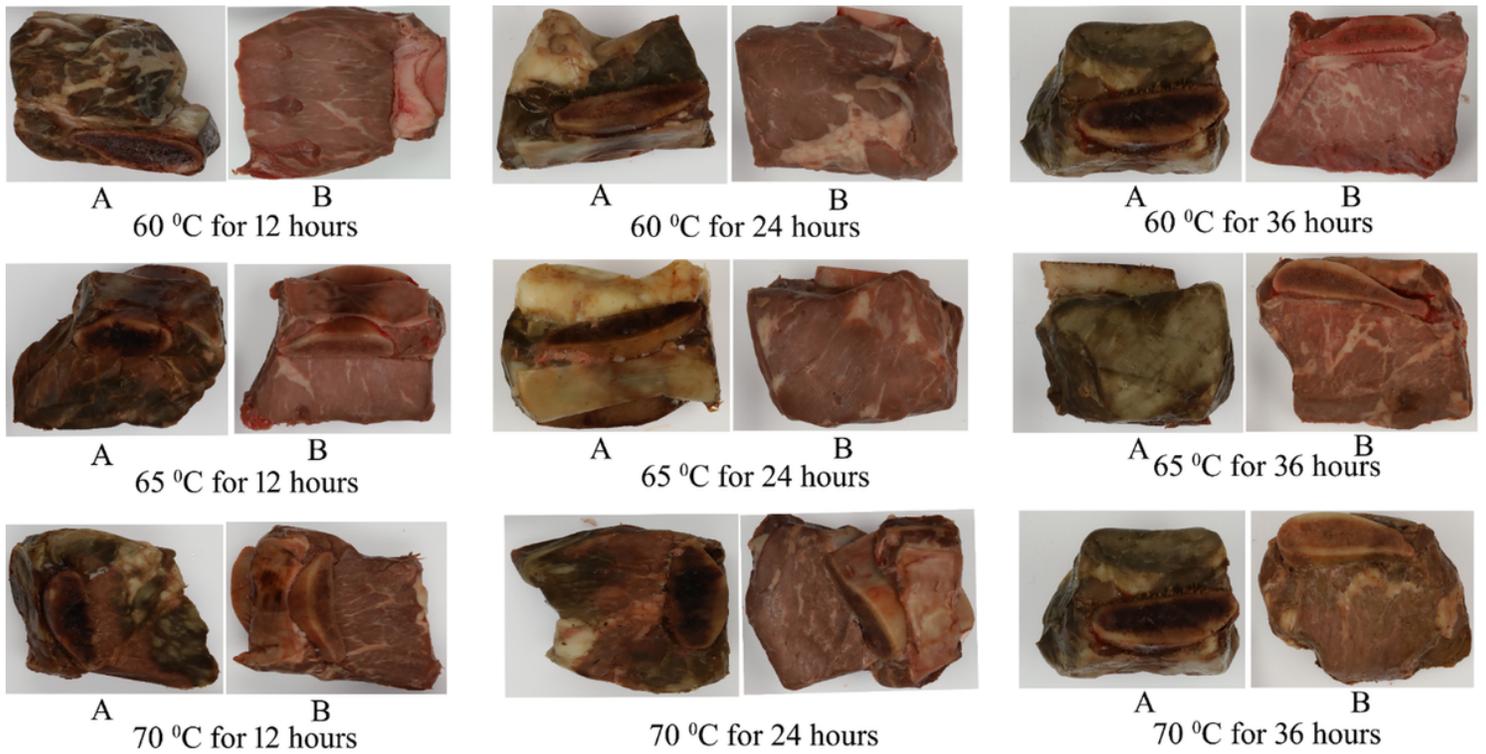


Figure 1

Effect of different packaging materials on colour of beef short ribs processed by sous vide at various temperature and time: (A) Polybag pouch, thickness 70 μm , tensile strength of 37-52 N/mm^2 , oxygen permeability (OTR) 50 $\text{cc}/\text{m}^2/\text{day}$ at 23 $^{\circ}\text{C}$, and water vapour transmission rate (WVTR) 8 $\text{g}/\text{m}^2/\text{day}$ at 38 $^{\circ}\text{C}$ and 90% relative humidity (RH); (B) Excel Aluminium foil pouch 3 layer laminate of polyethylene, ethylene vinyl alcohol and polyamide glued by food grade adhesives, thickness 79 μm , OTR $\leq 0.1 \text{ cc}/\text{m}^2/\text{day}$ at 23 $^{\circ}\text{C}$, 65% RH, and WVTR $\leq 0.1 \text{ g}/\text{m}^2/\text{day}$

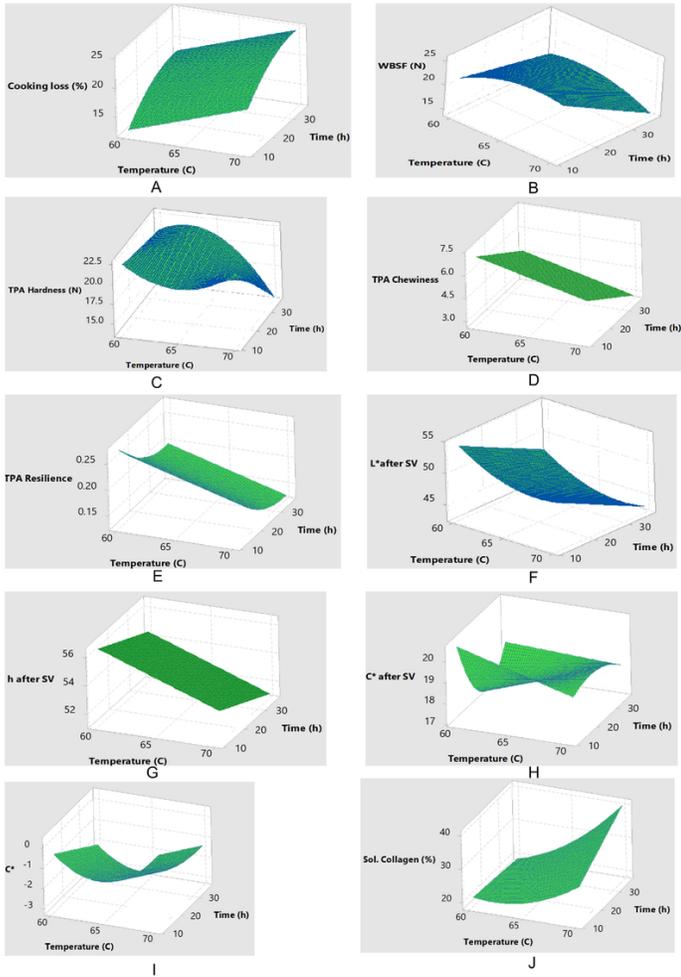


Figure 2

Surface plot of quality parameters at different sous vide temperature ($^{\circ}\text{C}$) and time (h) on physical parameters: (A) Cooking loss (%), (B) Warner Bratzler shear force (N), (C) TPA hardness (N), (D) TPA chewiness, (E) TPA resilience, (F) Lightness (L^*) after sous vide, (G) Hue angle (h) after sous vide, (H) Chroma (C^*) after sous vide, (I) Change in chroma (ΔC^*) after and before sous vide, (J) soluble collagen (%) after sous vide

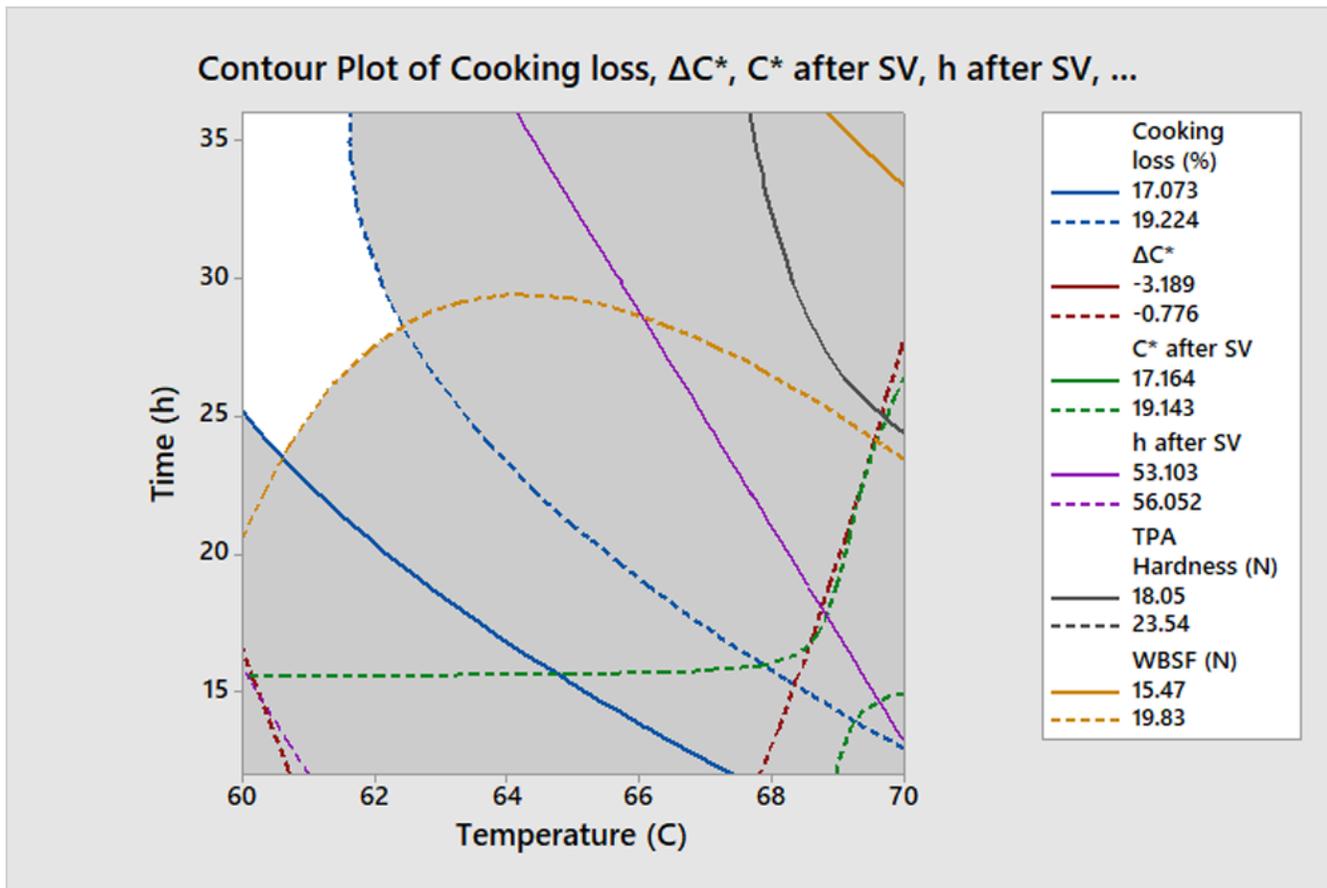


Figure 3

The overlaid contour plot showing effect of sous vide temperature and time on the lower and upper limit of cooking loss (%), Warner Bratzler shear force (WBSF), TPA hardness, hue angle and chroma. The white area covering the temperature range of 60 to 63 °C and time range of 24 to 36 h represents the optimum feasible area of sous vide processing.