FACTORS AFFECTING ACCEPTABILITY OF DRY-CURED HAM THROUGHOUT EXTENDED RIPENING UNDER "BODEGA" CONDITIONS

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Background

A great number of studies have been devoted to elucidate the relationship between several chemical parameters and different sensory traits in Iberian and other types of dry-cured hams. All these studies indicate these changes contribute an improvement in the quality of the cured ham. However, when these changes are very intense a decrease of the quality is observed, since they are responsible for the appearance of defective textures (soft and pasty to the touch, and exhibits extraneous aroma and flavour). Several studies have suggested that uncontrolled proteolytic activity results in considerably impaired protein mechanical properties.
Little research has been dedicated to determine the relationship among the instrumental measurements of texture and colour, biochemical processes, sensory parameters and acceptability in quality dry-cured hams of white breeds.

Objectives

The aim of this work was to study the evolution of chemical, color, and texture parameters to investigate their influence on different sensory characteristics, and to establish which of these sensory traits show a greater effect on the overall quality of dry-cured ham from white breeds. These would be useful for ham producers, since they would allow controlling those features directly related to the acceptability of the ham.

Materials and methods

Samples were 28 dry-cured hams (9-10 kg) which were obtained from pig breeds accepted by Denomination of Origin Teruel. All hams, used in this work, had been cured for 1 year and were purchased from various retail markets. The samples were stored at 18ºC and 75% relative humidity conditions from 12 months to 26 months. The central part of the ham was used for the analysis. The *pH* of dry-cured ham samples was measured using a micro pH meter model 2001 (Crison Instruments, Barcelona, Spain). *Lipid oxidation* was measured by the 2-thiobarbituric acid (TBA) method of Pfalzgraf (1995). TBARS values were expressed as mg malonaldehyde/kg sample. *The extraction of lipids* was made by the method of Folch et al. (1957). *Acidity* was made by the determination of percentage of oleic acid which was carried out by the method recommended by International Union of Pure and Applied Chemistry. Standard Methods for the Analysis of Oils, Fats and Soaps (1964). *Non protein nitrogen* was carried out by the method recommended by Norma ISO/R 937. The supernatant were analysed for nitrogen content following the AOAC (1984) Kjeldahl method. Nitrogen content in the supernatant, referred to 100 g meat, yielded total non protein nitrogen (NPN). *TVB content* was determined by steam-distillation according to the reference method (Decision 95/149/CE). Results were expressed as mg TVB-N per 100 g dry-cured ham. *Colour instrumental measurement*: a reflectance spectrophotometer (Minolta CM-2002; Osaka, Japan) was used to measure dry-cured ham colour at the surface of *Biceps femoris* and *Semimembranosus* muscles. The parameters registered were CIE L* (lightness), a* (redness), and b*(yellowness). *Texture analysis* was made in form of texture profile analysis (TPA) (Bourne, 1978; Henry et al., 1971) with a TA-XT2 Texture Analyser (Stable Micro Systems, Godalming, UK). From TPA curves, the following texture parameters were calculated: hardness, springiness, cohesiveness, adhesiveness, chewiness and gumminess. *Sensory Analysis:* **Trained panel:** The ham samples were sliced approximately 1,5 mm thickness. Each sample was presented at room temperature for evaluation. Intensities of aroma, flavour, tastes and texture factors were based on a structured scale (1-9). **Consumers**: Thirty consumers were recruited from students, faculty and staff of Faculty of Veterinary Science. Each Panellist evaluated samples (1.5 mm thickness) at home simulating common serving conditions. The hams were evaluated at 12, 14, 16, 18, 20, 22 and 26 months of storage. The score was based

on an acceptability scale of 9 points, where 1= very disgusting and 9= very pleasant. *Statistical analysis:* the effect of maturation-ripening time was carried out by analysis of variance, using the ANOVA, and Principal component analysis PCA procedure (SPSS, 11.5).

Results and discussion

During extended ripening some changes in *acceptability* were evident (Figure 1). The *consumer panel* did not find significant differences ($p>0.05$) in hams from 12 to 26 months (total manufacturing time), despite the fact that a trend to lower values was apparent from the 18th month. Acceptance scores were above 6. The *trained taste panel* found significant differences ($p<0.05$) after 22 months. A sharp decrease was evident at this point; after which scores were below 5.

All *chemical maturation parameters* increased throughout storage time, except pH (Figure 2). The parameters which showed a major variation were the intramuscular and subcutaneous acidity, NPN and TBA. They indicated an increasing ripeness and oxidation. In the last stage (22-26 months) a larger significant change ($p<0.05$) was observed in NPN and TBA indices, whereas acidity (lipolytic index) changed more rapidly during the first months of storage. These results supported the hypothesis of a large aroma development within the first stage, whereas it became stable at the end of storage.

Figure 3 (a, b) shows the changes in the *instrumental parameters of texture*. There were evident changes in *Semimembranosus* muscle, in particular regarding hardness which reached significant higher values from the 18th month. Adhesiveness decreased, but increased significantly ($p<0.05$) at the end of storage. The same changes were observed in *Biceps femoris* muscle though they were not so remarkable.

Figure 4 shows the results of the *visual and olfactive sensory parameters.* Colour of *Biceps femoris* increased significantly ($p<0.05$). Aroma reached a maximum peak at the 18th month, then decreased probably it due to the evaporation of the volatile aromatic substances. With regard to *texture parameters* (Figure 5) crumbliness showed a significant decrease ($p<0.05$) from the 18 month of storage, whereas pastinesss and adhesiveness strongly increased ($p<0.05$) from the 18-20 months to the end of storage.

The evolution of *flavour* (Figures 6) was very similar to that of aroma; it showed no significant variation ($p>0.05$) after 20 months of storage. *Rancidity* decreased significantly ($p<0.05$) at the end of storage.

Figure 7 is a plot of the principal components analysis loadings for the first two partial least squares of components.The first component was able to predict 54.41 % of the variation of the whole study. Acceptability, crumbliness and cohesiveness reached a high negative loading on component 1, whereas chemical parameters of maturation had a positive loading on component 1. The second component explained only 18.33 % of the variation. Positive loadings had *Biceps femoris* cohesiveness and some of the colour parameters. The negative coefficients on component 2 turned out to be the pastiness, adhesiveness and the rest of the colour parameters.

Following PCA, 12-14-month-ripened hams were in the left quadrant with negative coefficients on the component 1. As storage time and ripeness increased they moved to the left quadrant with positive values in the component 2, and thereafter to the upper-right quadrant. The final trend was towards the lower-right quadrant, with positive a coefficient on the component 1 and a highly negative on the component 2. It must be emphasized that adhesiveness and pastiness were located in this same quadrant.

Conclusions

Results demonstrated that ham acceptability showed no significant differences ($p>0.05$) from 12 to 20 months (total manufacturing time), while it decreased significantly ($p<0.05$) at 22-26 months. Principal component analysis of all data brought about a comprehensive explanation of the biochemical, instrumental and sensory parameters involved in the acceptability decrease. In fact, high pastiness and adhesiveness values, as measured by both sensory and instrumental methods, appeared to be most correlated with decreasing acceptability. Those were the result of an excessive proteolysis, as revealed by biochemical maturation indices.

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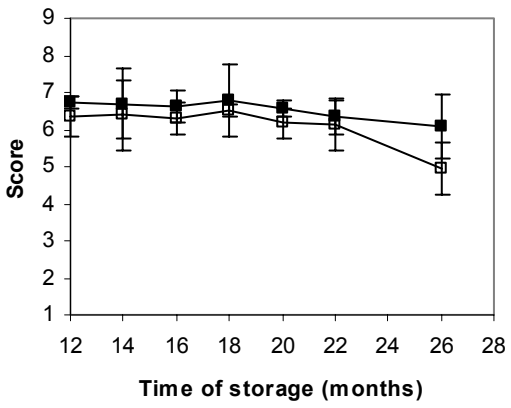

Fig.1. Evolution of the acceptability during time of storage. (■) Consumer's acceptability, (□) Trained Panel's acceptability.

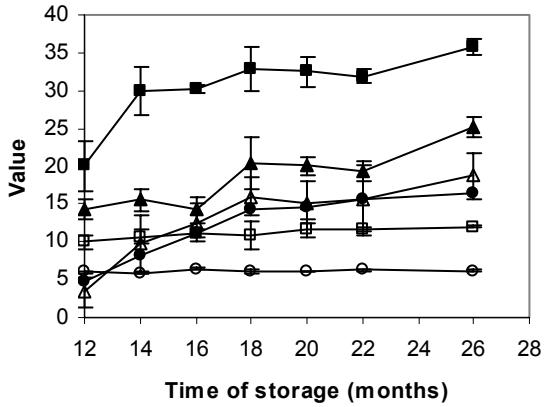

Fig.2. Evolution of chemical parameters during time of storage. (■), TBA x 100 (mg malonaldehído/kg), (□)TVB-N /10 (mg TVB-N/100 g), (▲)NPN/10 (mg NPN/100 g), (Δ) Intramuscular Acidity (% ac. Oleico), (●) Subcutaneous Acidity (% ac. Oleico), (○) pH.

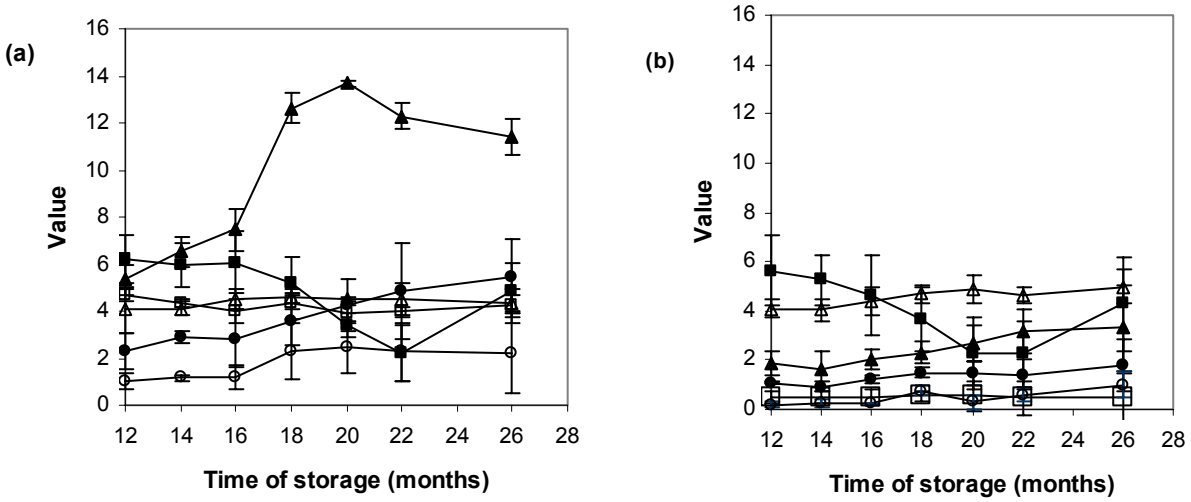

Fig.3. Evolution of instrumental texture parameters during time of storage. (a) *Semimembranosus*, (b) *Biceps femoris*; (■) Adhesiveness/-10 (gs), (□) Cohesiveness x 10 (g), (▲) Hardness/1000 (g), (Δ) Springiness x 10, (●) Gumminess/1000, (○) Chewiness/1000.

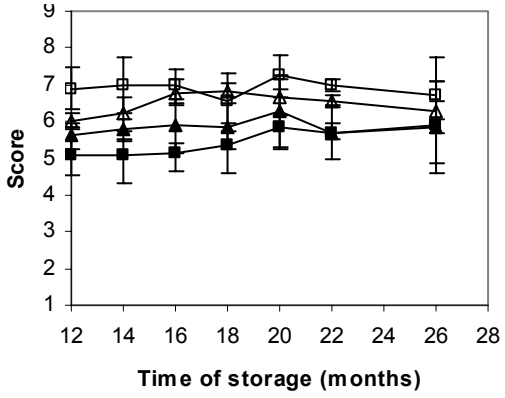

Fig.4. Evolution of sensory analysis parameters of aroma and colour during time of storage. (■)Colour of cured in *Biceps femoris*, (□) Colour of cured in *Semimembranosus*, (▲) Colour homogeneity, (Δ)

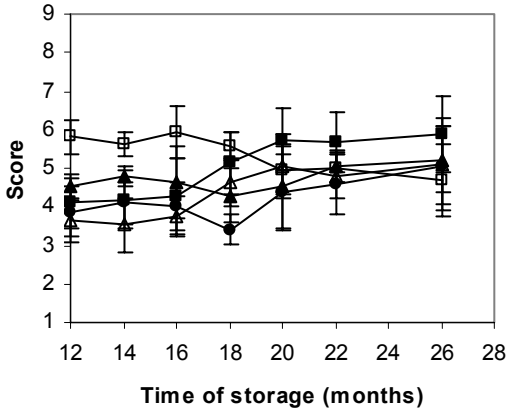

Fig.5. Evolution of sensory analysis parameters of texture during time of storage. (■) Hardeness, (□) Crumbliness, (▲) Pastiness, (Δ) Fibrousness, (●), Adhesiveness.

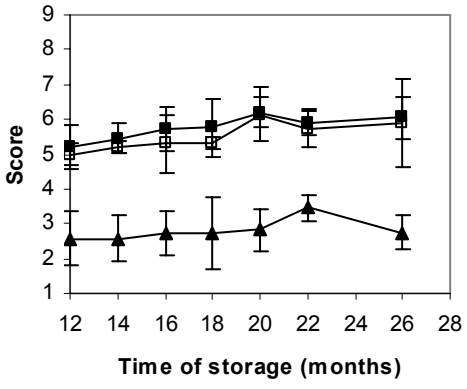

Fig.6. Evolution of sensory analysis parameters of flavour during time of storage. (■) Ham flavour, (□) Saltiness, (▲) Rancidity flavour.

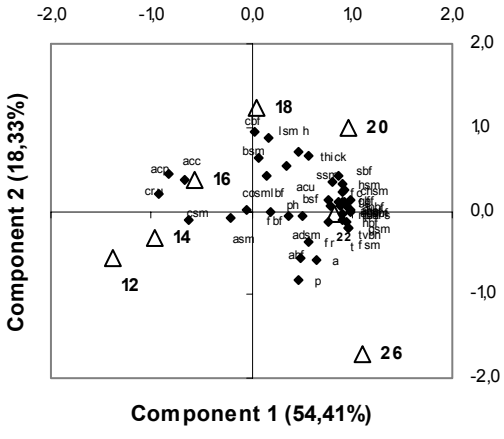

Fig.7. Biplot for the first two principal components. (Δ) centroid for hams at each storage time,(♦)studied parameters

EFFECT OF WALNUT, MICROBIAL TRANSGLUTAMINASE AND STORAGE TIME ON THE WATER AND FAT BINDING CAPACITY OF SALT-FREE BEEF BATTERS

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Background

Consumers are currently more health conscious and as a result demand healthier products. Sodium chloride intake has thus been associated with arterial hypertension and animal fat consumption with cardiovascular disease, one of the main causes of death in industrialised countries. Accordingly, current recommendations aim to reduce daily sodium and saturated fatty acid intake. One way of doing this is to reformulate meat products by reducing the salt content and incorporating fatty acids with proven health benefits.
Different studies have shown that the frequent consumption of walnuts reduces the risk of cardiovascular disease, since it lowers serum cholesterol and favourably modifies the lipoprotein profile (Sabaté, 1993). One way of promoting walnut intake would be to include it as an ingredient in frequently consumed foods, such as meat derivatives, which can thus be made to incorporate health benefits.
In meat products, salt (NaCl) extracts the myofibrillar proteins which are mainly responsible for the heat-induced development of a functional protein matrix by immobilising water (Acton *et al*., 1985). Salt reduction in most products will have an adverse effect on protein extraction, water and fat binding properties and gel strength, and will also alter palatability attributes (Girard *et al*., 1990). In an attempt to reduce the salt level, microbial transglutaminase (MTG) has been increasingly used as a cold meat binder in the absence of salt by combining it with suitable food protein (Kuraishi *et al*., 1997) and crosslinking meat proteins. However, there are hardly any studies on how the storage time in chilling (real commercial conditions) affects the characteristics of raw meat products prepared with MTG as a cold-set binder.

Objectives

The aim of this work, using response surface methodology, was to test how the addition of increasing amounts of walnut (W), microbial transglutaminase/sodium caseinate (MTG/C) and the storage time (ST) (up to 11 days) at 3 °C affect the water and fat binding properties of beef batters formulated in the absence of salt and phosphates.

Materials and methods

Post-rigor beef *semimenbranosus* (top round) was purchased from a local market, ground through a 6 mm plate and divided into 20 (400 g) batches, vacuum packed and stored at -20° C until use. Additives used for preparing meat batters included walnut, supplied by La Morella Nuts, S.A. (Tarragona, Spain), (particle size of 12μ) and microbial transglutaminase/caseinate (ACTIVA EB, Ajinomoto Europe Sales GmbH, Germany) in a formulation containing sodium caseinate 60%, maltodextrin 39.5% and transglutaminase 0.5%. Transglutaminase activity was approximately 34-65 units/g. In all the formulations the meat protein content was adjusted to a constant level (18%) with water, although the total protein content was dependent on the proportion of added walnut.

Beef meat, MTG/C (dissolved in added cold water) and walnut were homogenized in a cutter (Stephan Universal Machine UM5, Stephan U. Stephan U. Söhne GmH & Co., Hameln, Germany) under chilled vacuum conditions. Preparation of the meat batters took 3 min., during which time the final chopping temperature did not exceed 12 °C. Immediately after the homogenate preparation, the batters were stuffed into plastic tubes (diam 3.4 cm) and hermetically sealed. The homogenate samples were then stored in a cold room at 3 °C at various storage times (ST) shown in Table 1.
After the storage time required in a cold room, the meat batters in the plastic tubes were heated (30 min. 70 ° C) in a waterbath. After heating, the containers were opened and left to stand upside down (for 40 min.) to release the exudate. Total loss (TL) was expressed as a % of initial sample weight. Water loss (WL) was

determined as a % weight loss after heating the total loss (TL) for 16 h on a stove at 100 °C. Fat loss (FL) was calculated as the difference between TL and WL. Determinations were carried out in quintuplicate.
Response surface methodology (RSM) was used to study the simultaneous effect of the three experimental variables. Experimental design and statistical analysis were performed using Statgraphics plus 2.1 (STSC Inc., Rockville, MD). Five levels of each variable were chosen following the principles of the central composite design principle (Khuri and Cornell, 1996). The levels of the variables are shown in Table 1.

Results and discussion

Water and fat binding properties
Analysis of variance indicated that among the regression models for the water and fat binding properties of the salt-free beef batters with varying levels of walnut and MTG/C at different storage times, only the model for total loss (TL) was significant (Table 2), while the models for water loss (WL) and fat loss (FL) were not significant. Furthermore, the absence of any interaction effects on total loss indicates that the effect exerted by each variable was not affected by the others. The effect of the variables studied is shown in Fig. 1.
The level of the added walnut has a negative linear effect, the greater the amount of walnut in the meat batters the lower the total loss (TL) values (Fig. 1A, C). This behaviour could be due to the lower water content present in the samples with greater walnut content, since the incorporation of the walnut is at the expense of water, with the concentration of the meat protein remaining constant. Therefore, for the same concentration of meat protein, the samples with greater water content will lose more than the samples with a lower water content and a greater amount of walnut, since the water content of the walnut is very low.
The incorporation of increasing amounts of MTG/C does not seem to affect the amount of TL during heating (Fig. 1A, B). The apparently high results of total loss in this experiment in salt-free beef batters without walnut are agreed with the low ionic strength in the formulation, which reduced myofibrillar protein extraction and therefore increased cooking losses. These results are the same as those obtained by other authors in restructured meat prepared with MTG and without salt (Kerry, 1999). Although some authors suggest that non-meat proteins like caseinate with MTG are necessary to ensure proper binding properties, in the conditions assayed in this study, meat batters were obtained with poor water and fat binding properties. There are apparently contradictory results about the effect of MTG on water and fat binding properties and these differences could be due to the level and different type of MTG added and the conditions in which they are used (time and reaction temperature; presence of other ingredients such a salt, phosphates, and non-meat proteins, species of origin, etc.) (Pietrasik and Li-Chan, 2002). The storage time in chilling did not have any significant effect ($P > 0.05$) on total loss.

Conclusions

The results obtained in this study suggest that the gel/emulsion systems induced during the heating process prepared with MTG/C in the absence of salt and phosphates form structures with poor binding properties. The detrimental effects caused by the absence of salt in beef batters were not overcome by the addition of MTG/C used in this study. However, the presence of walnut in salt-free beef batters made with MTG/C reduces cooking losses, thereby improving water and fat binding properties.

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Table 1. Levels of variables established according to the central composite rotatable design.

Treatment	Walnut %	MTG %	Storage time (days)
1	12.50	1.00	3
2	3.20	1.00	9
3	15.67	0.65	6
4	12.50	0.30	9
5	12.55	0.30	3
6	7.85	0.65	6
7	0	0.65	6
8	7.85	0.65	6
9	7.85	0	6
10	3.20	0.30	3
11	3.20	1.00	3
12	7.85	0.65	1
13	3.20	0.30	9
14	7.85	1.24	6
15	12.50	1.00	9
16	7.85	0.65	11
17	7.85	0.65	6
18	7.85	0.65	6
19	7.85	0.65	6
20	7.85	0.65	6

Table 2. Analysis of variance of the regression models for water and fat binding properties of salt-free beef batters.

	MODEL[a]	R^{2b}
Water and fat binding properties		
TL	0.023 (*)	77.5 – 57.39
WL	NS	
FL	NS	

[a] * = significant at P<0.05; NS = not significant.
[b] Fitted for degrees of freedom.

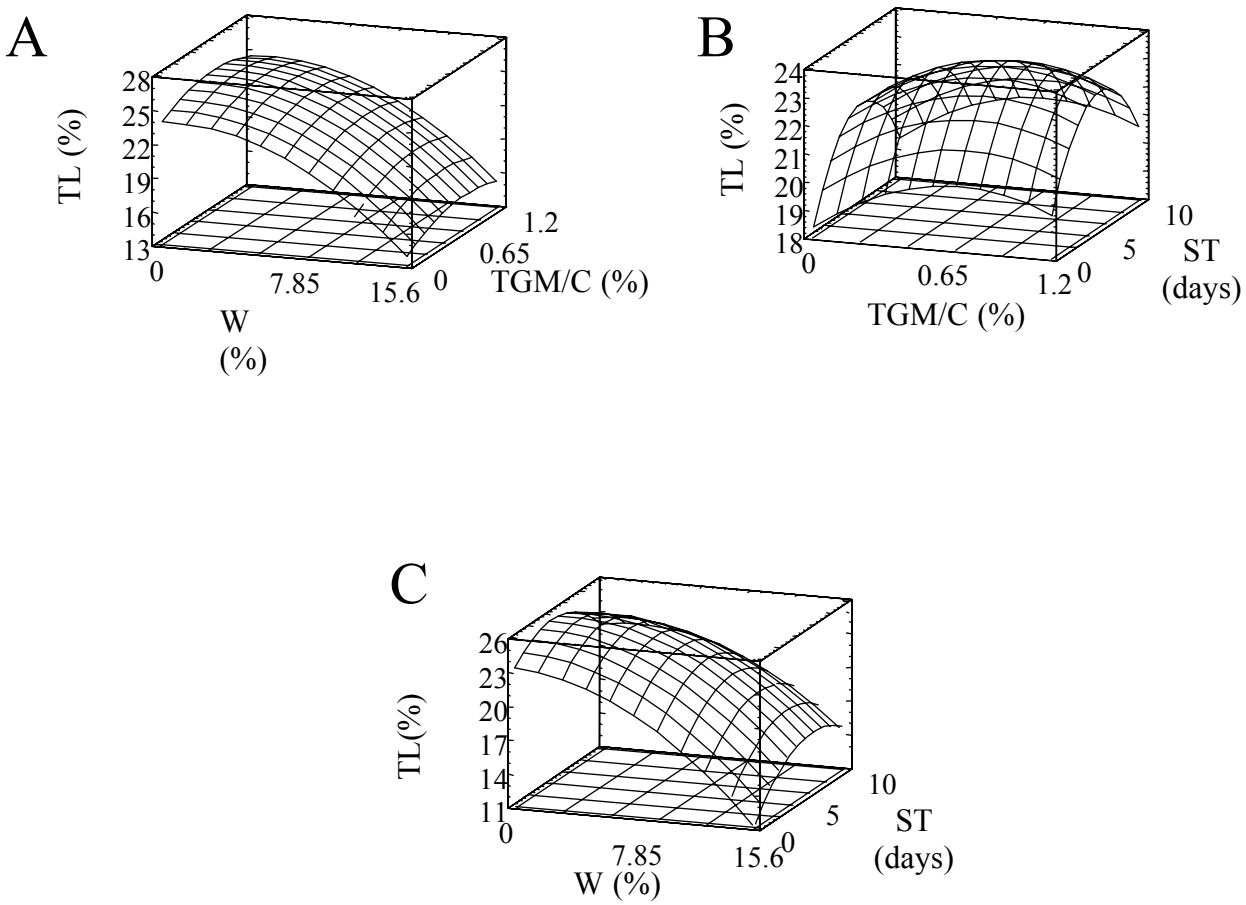

Figure 1. Effect of walnut (W), microbial transglutaminase/sodium caseinate (MTG/C) and storage time (ST) on total loss (TL) of salt-free beef meat batters. (A) W and MTG/C at 6 days (ST); (B) MTG/C and ST at 7.85% W; (C) W and ST at 0.65% MTG/C.

ADDITION OF WHEY PROTEIN CONCENTRATE AND SODIUM CHLORIDE TO BEEF MUSCLES: EFFECTS OF TUMBLING PROCEDURES AND *SOUS VIDE* COOKING TREATMENT ON TECHNOLOGICAL PARAMETERS, PHYSICAL PROPERTIES AND SENSORY QUALITY

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Background

Sous vide processing is the application of a cooking-pasteurization thermal process to food packaged in a hermetically sealed vacuum pouch or tray (Church & Parsons, 1993). Advantages associated with *sous vide* processing include: a superior flavour profile compared to conventionally prepared food, increased tenderness and moisture, improved colour retention and reduced nutritional loss. Furthermore, processing and storage in evacuated pouch increase product shelf life by inhibiting microbiological and chemical spoilage (Vaudagna *et al*., 2002). The *sous vide* cooked beef muscle is an interesting alternative to diversify the marketing of meat, however, this type of products present technological difficulties such as the retention -inside the packaging- of the juice lost during processing (Church & Parsons, 2000). This problem is relevant, particularly regarding commercial profit and preservation of the sensorial and nutritional characteristics of the product. In order to increase water holding capacity (WHC) of meats and consequently reduce cooking weight loss, it has been suggested the use of mild thermal treatments (Vaudagna *et al.*, 2002), the addition of salts, such as sodium chloride (SC) and/or alkaline phosphates (Sofos, 1989), and more recently, the use of natural functional ingredients, i.e. whey protein isolate (WPI) and concentrate (WPC), soy protein isolate, polysaccharide gums, starches, blood plasma, etc. (Chen & Trout, 1991). The higher viscosity of the brines containing those natural ingredients resulted in a non homogenous brine distribution with detrimental consequences for WHC, cooking weight loss and sensory quality. This is particularly relevant in high quality products processed with low injection rates (10 % - 20%) where concentrated brines are used. Consequently, to improve brine distribution, the injection and tumbling procedures become critical. Mechanical treatment of meat tissue by tumbling (massaging) is well recognized and accepted techniques in the meat industry (Xargayo & Lagares, 1992). However, little information is available regarding the effect of extended tumbling procedures on binding and sensory quality of cooked beef pieces (Pietrasik & Shand, 2004), particularly when natural functional ingredients are added to them. The combination of pre- and post- injection tumbling procedures may provide a useful means to improve brine distribution, water binding properties and sensory quality.

Objectives

To evaluate the combined effect of two tumbling procedures (pre- and post- injection step) on technological parameters, physical properties and sensory quality of WPC + SC added beef muscles cooked using the *sous vide* system.

Materials and methods

A 3 x 3 factorial design with three replicates was applied in this experiment. The major variables investigated were tumbling treatments prior to injection step (pre-injection tumbling, PreIT) and after injection step (post-injection tumbling, PostIT), see Table 1. For the experimental design, fifty four *Semitendinosus* muscles were dissected from British breed steer carcasses 48 h post slaughter, trimmed free of fat, vacuum packaged (Cryovac BB4L, Sealed Air Co., Buenos Aires, Argentina), stored for 72 h at 1.5 ± 0.5°C and then processed. The trimmed raw muscles had an average weight of 1579.8 ± 166.1 g, and an average pH of 5.62 ± 0.08. Then, two procedures were carried out, by one hand, the muscles selected for PreIT were weighed, vacuum packaged in proper bags (Cryovac CN510, Sealed Air Co., Buenos Aires,

Argentina) and continually tumbled (2.5 rpm) for 1.5 or 3 h at 1.5 ± 0.5 °C in a Lance Industries tumbler (model LT-15, Allenton, USA) using a drum load of 45 kg (half of working capacity). On the other hand, the muscles non tumbled (NPreIT) were weighed, vacuum packaged in the bags previously described and stored at 1.5 ± 0.5 °C until further injection. Then, the PreIT and NPreIT muscles were weighed and injected using a hand operated brine pump (Dick Lokespritze Esslingen A.NX., Germany). Brine was formulated to give a concentration of 3.5 %WPC (Lacprodan 80, Arla Food Ingredients S.A., Buenos Aires, Argentina) and 0.7% SC (Dos Anclas, Buenos Aires, Argentina) in the injected product, and it was added at a rate of 20% (w/w). After injection, two procedures were applied, by one hand, non tumbled muscles (NPostIT) were vacuum packaged in proper bags (Cryovac CN510, Sealed Air Co., Buenos Aires-Argentina) and then refrigerated overnight at 1.5 ± 0.5°C until *sous vide* cooking was performed. On the other hand, the muscles selected for PostIT were vacuum packaged in the Cryovac CN510 bags, continually tumbled (5 rpm) for 2 or 10 h at 1.5 ±0.5°C in the equipment previously described and stored overnight at 1.5 ± 0.5 °C until *sous vide* cooking was carried out. Then, all muscles were weighed, vacuum packaged into cook-in bags (Cryovac CN510, Sealed Air Co., Buenos Aires, Argentina) and cooked in a water cascading retort (Microflow Barriquand, Roanne, France) in batches of twenty four muscles (the device was operated in static basket mode). Time-temperature evolutions in the slowest heating point (SHP) of three muscles and in the retort chamber were measured using a T type thermocouple and recorded with a digital multimeter Hydra 2625A date logger (John Fluke Mfg. Co., Inc., Everett, USA). In the present study, a processing temperature and time combination of 70°C – 2 min were applied at the muscle SHP. This treatment has been suggested in order to achieve a 6D reduction of *Listeria monocytogenes* (FAIR CT96-1020, 1999). Immediately after the heat treatment, samples were immersed in an ice-water bath until the temperature at SHP reached 10 °C and then were stored at 1.5±0.5 °C for 18 h until further testing.

The technological parameters measured on each muscle were PreIT weight loss percentage (P_1), PostIT weight loss percentage (P_2), cooking-pasteurisation weight loss percentage (P_3) and total weight loss percentage (P_T). Each weigh loss percentage was determined using the relationship $P_i = 100 \times (m_i - m_f)/m_{trm}$, where, for P_1: m_i is the mass of the trimmed raw muscle (non injected) and m_f is the mass of the muscle after PreIT treatment. For P_2, m_i is the mass of the injected muscle and m_f is the mass of the muscle after PostIT treatment, while for P_3, m_i is the mass of muscle after PostIT treatment and m_f is the mass of the muscle after thermal process. Finally, for P_T, m_i is the mass of the trimmed raw muscle (non injected) and m_f is the mass of the muscle after thermal process. All percentages were based on the weight of the trimmed raw muscle (non injected), m_{trm}. The pH values of raw and cooked muscles were measured in duplicate on an homogenate (5 g of sample: 25 ml of distilled water buffered at pH 7) with a pH-meter (Thermo Orion 710A+, Beverly MA, USA) equipped with a combination pH electrode (Thermo Orion Model 8102BN ROSS Electrode). Warner Bratzler shear (WBS) was determined on ten cylinders (3 cm height; 1.27 cm in diameter) obtained from 2.0 cm slice located in the muscle medial portion. For this purpose, Warner–Bratzler meat shear device (Chatllon, New York, USA) with a triangular shear was used. Also, visual appearance of 1.5 cm slice also separated from the muscle medial portion was judged by ten trained panellists using a Veri-Vide CAC120 box (illuminant D_{65}). A seven-point scale was used for cooked beef colour, and a five-point scale for colour uniformity, following the specification of AMSA (1991). The level of defects in the slices was evaluated by a five-point scale of 1=*None*, 5= *Extreme*. The amount of defects (percentage by slice area) was also evaluated using a seven-point scales of 1= *No defect* (0%), 7= *Total* (100%). For statistical analysis, data were analysed as a 3 x 3 factorial design with PreIT and PostIT treatments as main effects (A and B, respectively). All data were analysed by SPSS (Statistical Package for the Social Sciences) 12.0 for Windows (SPSS Inc., Chicago, IL, USA). The Tukey's multiple range test at P=0.05 was used to determine differences between means. Principal component analysis (PCA) was also performed with SPSS software.

Results and discussion

Neither PreIT nor PostIT treatments had significant effect ($P > 0.05$) on the pH of cooked muscles (injected or non injected). However, a significant pH increase was observed ($P<0.05$) for the injected + cooked muscles (pH = 6.11 ± 0.09) in comparison to, raw muscles (pH = 5.61 ± 0.08) and non injected + cooked (pH= 5.87 ± 0.07) ones. An overview of Figure 1 and Table 2, showed that the highest weight losses correspond to cooking-pasteurisation step (P_3). In addition, PostIT weight losses (P_2-Table 2) were higher than the PreIT ones (P_1-figure 1), probably as consequence of the more severe PostIT procedure. In Fig. 1 it is also observed that increasing PreIT times up to 3 h had non significant effect on P_1 parameter ($P > 0.05$),

which values were in the range of 1.0-1.4%. In the present study, PreIT treatments were applied in order to provide more suitable physical conditions for the injection procedure. These treatments were suggested considering both, the high viscosity of the brines used and the higher resistance of beef to brine addition compared to pork and poultry (Pietrasik & Shand, 2004). According to Table 2, P_2, P_3 and P_T parameters were not significantly affected by PreIT times ($P > 0.05$) but they were markedly increased ($P < 0.05$) by PostIT times (Table 2). Then, PostIT time up to 2 h had not effect on them, but the extending of PostIT time to 10 h increased significantly these variables (Table 2). None of the evaluated weight losses was affected by interactive effects between PreIT and PostIT treatments (P=0.1341, P=0.3068 and P=0.4642, for P_2, P_3 and P_T respectively). As it was expected, PostIT weight loss (P_2) corresponding to a tumbling time of 10 h was higher than those one from muscles, either tumbled by 2 h or non tumbled (NPostIT). This result could be due to a larger tissue disruption induced by the extended tumbling time. Also, muscles treated by PostIT of 10 h presented a cooking-pasteurisation weigh loss (P_3) higher than those muscles processed for 2 h or NPostIT. This effect is different from the one observed in muscles added of conventional functional ingredients, in which the extended tumbling has been described as a factor that improves the water binding characteristics and reduces purge and cooking losses. Generally, extended tumbling is required to incorporate brine into the muscle cells and to provide more suitable conditions for protein solubilization and extraction (Xargayo & Lagares, 1992; Pietrasik & Shand, 2004). The described discrepancy can be attributed to differences in formulation (WPC and SC ingredients), in injection levels, in meat composition and structure, and in cooking procedure (heating rate, temperature and time).Even though the injected muscles processed by PreIT and PostIT treatments have P_T in the range of 5 – 10 % (Table 2), these are significantly lower ($P < 0.05$) than those of non injected muscles similarly processed (25-30%, data not shown). It proved that WPC and SC addition successfully reduced weight losses between 2-3 times. As can also be seen in Table 2, neither PreIT nor PostIT treatments had significant effect ($P > 0.05$) on the instrumental tenderness (WBS) of injected + cooked muscles. However, it was detected a trend to increase tenderness in muscles treated by PostIT for 10 h. A correlation between WBS values and a hedonic scale, let us classified injected + cooked muscles as "very tender".

In Figure 2, P_2, P_3 and P_T weight losses, sensory ("cooked beef colour", "colour uniformity", "level of defects") and instrumental (WBS) parameters, and NPostIT and PostIT treatments have been plotted as it was obtained by PCA. The first and second axes (PC1 and PC2) describe 62.3 and 19.8 % of overall variation, respectively. The horizontal axis (PC1) discriminates treatments by the three weight losses (left side) and sensory and instrumental parameters (right side). Even though muscles processed by the PostIT time of 10 h had higher weight losses than those of the muscles submitted to other treatments (NPostIT and PostIT of 2 h), they showed a more uniform *pinkish-gray* colour (3= s*mall amount of variation*) and improved tenderness (lower WBS values). Since the descriptor "level of defects" presented similar loadings for both components (0.65 and 0.66 for PC1 and PC2, respectively), then this sensory parameter would also be related to PC2 axis. Thus, Fig. 2 shows that muscles without PostIT (NPostIT) or processed by a PostIT time of 2 h exhibited a higher "level of defects" than those treated by a PostIT time of 10 h (this descriptor was evaluated as s*mall* and involved 20 to 40% of slices area). Taking into account present results, the PostIT time of 10 h seemed to be the most appropriate treatment to improve quality characteristics.

Conclusions

Contrary to our expectations, tumbling procedure applied prior to the injection one did not affect either technological parameters (tumbling and cooking weight losses) or sensory quality (tenderness and visual appearance) of cooked beef muscles. As well, a post injection tumbling time of 2 h had not influence on these parameters. However, extending post injection tumbling times to 10 h, improved brine distribution and sensory quality (cooked beef colour, colour uniformity, level and amount of defects and tenderness). The drawback was that tumbling and cooking weight losses were increased by this treatment.

Acknowledgements

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TABLE 1: Description of tumbling procedures applied.

PRE-INJECTION TUMBLING (PreIT)	POST-INJECTION TUMBLING (PostIT)
NPreIT (*)	NPostIT (**)
NPreIT (*)	5 rpm – 2 h
NPreIT (*)	5 rpm – 10 h
2.5 rpm – 1.5 h	NPostIT (**)
2.5 rpm – 1.5 h	5 rpm – 2 h
2.5 rpm – 1.5 h	5 rpm – 10 h
2.5 rpm – 3 h	NPostIT (**)
2.5 rpm – 3 h	5 rpm – 2 h
2.5 rpm – 3 h	5 rpm – 10 h

(*) PreIT non carried out. (**) PostIT non carried out.

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Figure 1: Percentage of pre-injection tumbling (PreIT) weight loss at the different PreIT times

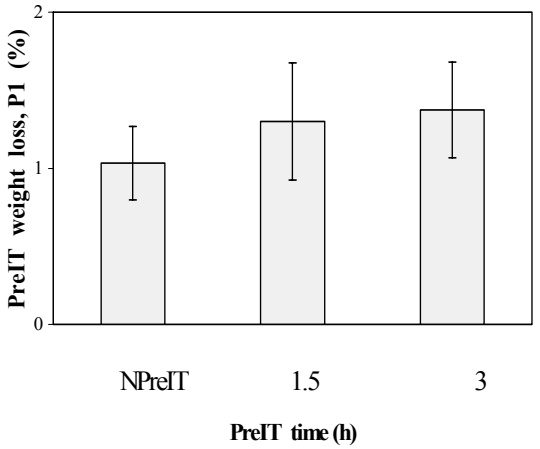
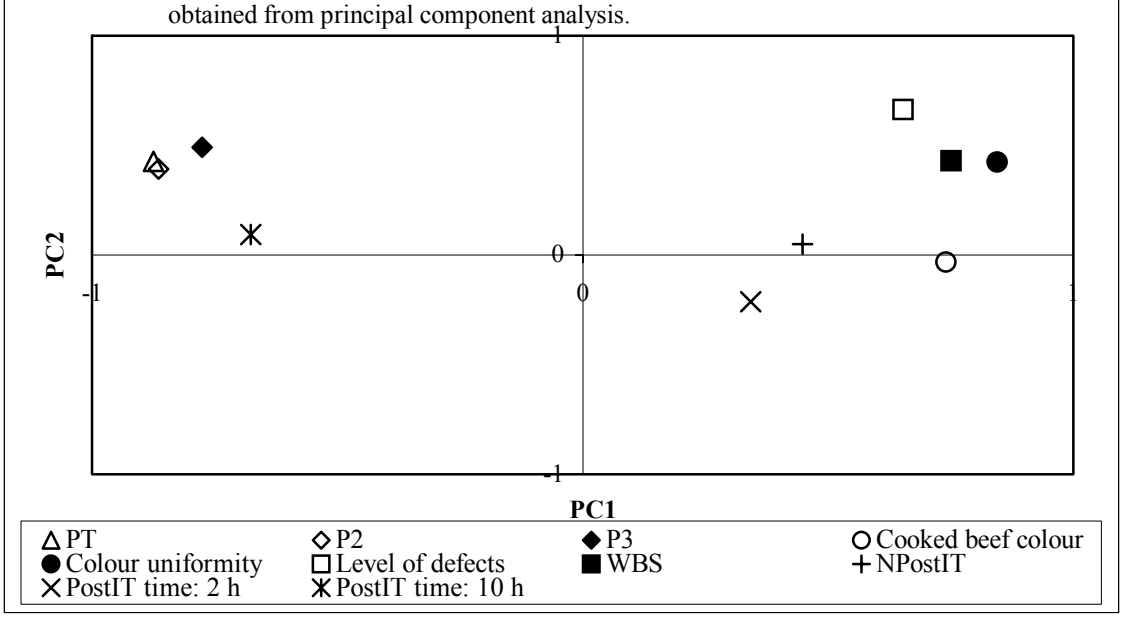


TABLE 2: Technological parameters and Warner Bratzler Shear (WBS) values of muscles processed by the different pre- and post tumbling procedures.

FACTORIAL MAIN EFFECT AND INTERACTION		TECHNOLOGICAL PARAMETERS			WBS
		P_2 (%)	P_3 (%)	P_T (%)	(lb in^{-2})
A: PreIT	NPreIT	4.2 ± 1.0	21.8 ± 1.6	7.3 ± 2.4	5.0 ± 0.2
	2.5 rpm – 1.5 h	3.9 ± 1.0	21.4 ± 1.1	6.7 ± 2.2	4.8 ± 0.1
	2.5 rpm – 3 h	4.2 ± 1.1	21.8 ± 1.8	7.6 ± 2.7	4.8 ± 0.1
	P-value	0.6800	0.6900	0.6609	> 0.05
B: PostIT	NPostIT	3.8 ± 0.8 **b**	21.0 ± 1.0 **b**	6.3 ± 1.3 **b**	4.9 ± 0.1
	5 rpm – 2 h	3.5 ± 0.8 **b**	21.1 ± 1.1 **b**	5.9 ± 1.7 **b**	5.1 ± 0.1
	5 rpm – 10 h	4.9 ± 0.8 **a**	22.9 ± 1.5 **a**	9.4 ± 2.3 **a**	4.7 ± 0.2
	P-value	0.0021	0.0066	0.0020	> 0.05
Interaction A x B	*P-value*	0.1341	0.3068	0.4642	> 0.05

Means with different letters in the same column (within each main effect, A or B) are significantly different ($P < 0.05$)

Figure 2: Loadings of technological, instrumental and sensory parameters and post-injection tumbling (PostIT) treatments on the first two components (PC1 vs PC2) obtained from principal component analysis.



OPTIMISING THE MEASURING OF MEAT FLAVOURS BY MEANS OF AN ELECTRONIC NOSE VIA SENSOR SELECTION

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Background

A reproducible method for detecting flavours is an important precondition for the successful employment of the electronic nose when evaluating the quality of foodstuffs. The results of the executed measuring of meat flavours showed a number of factors which are to be optimised to obtain reliable results when using gas sensor arrays (Rosenbauer *et al*. 1998). The precise knowledge of the sensor selectivity, i.e. what the signals of individual sensors contribute to the differentiation, apart from the factors examined by us, like the reference air temperature and humidity, the mass flow of the carrier gas, the sample volume, the surface, the incubation period and temperature of the sample is essential for the distinctiveness of a sensor system (Dederer and Troeger 2000).

Objectives

Subsequently, the possibility to improve the efficiency of the sensor system VOCmeter (Messrs. Motech in Reutlingen) was demonstrated by selecting the appropriate sensors and by optimising the evaluation of the sensor signals by means of a practical example (comparison of beef samples with different pH values during ageing).

Materials and methods

The employed system works on the principle of mixed sensors: It contains 4 metal oxide (MOX) and 8 quartz microbalance sensors (QMB).
The sample material was beef meat (*M. longissimus dorsi*) with a pH value of 5.5 and 5.9. The samples were stored in multilayer plastic bags at +2 °C for a total of 5 weeks. In each storage week, the samples were measured in a dynamic measuring modus under the following experimental conditions: Sample volume 10 g; thermostatisation of the samples by 35 °C for 30 minutes; carrier gas – purified air with 55 % humidity (related to 30°C); flow rate of the reference air 25 ml/sec; equilibration time of the sensors 120 sec; measuring period 5 min. Parallel to their measuring by means of the electronic nose, the meat samples were evaluated sensorial according to the criteria of flavour and tenderness on the basis of a 6-point scale.

Results and discussion

To establish the sensor rate relevant for differentiating the ageing condition of the beef samples, the sensor signals of the 4 MOX sensors and the 8 QMB sensors were evaluated separately. Particularly those sensors mainly reacting on polar substances and on common hydrocarbons, showed a significant increase in the signal intensity during the meat ageing.

The Figure 1 and 2 show the mean value of the maximum of the sensor reactions for beef samples with different pH-value and storage period. In case of MOX sensors, slight differences were established in the signal intensity between all samples stored for one week and an increasing signal intensity of all sensors after the second storage week. The highest signals were established for the samples with the pH-value 5.9. In this connection, sensor MT-J20 particularly reacting on polar substances and sensor MT-J0 which reacts on common hydrocarbons showed a significant increase of the signal intensity during meat ageing. The sensor indicated by the manufacture selectively for methane and permanent gases, reacted the least on the flavour differences. Only two of the eight QMB sensors reacted on the ageing-related differences of the meat flavour.

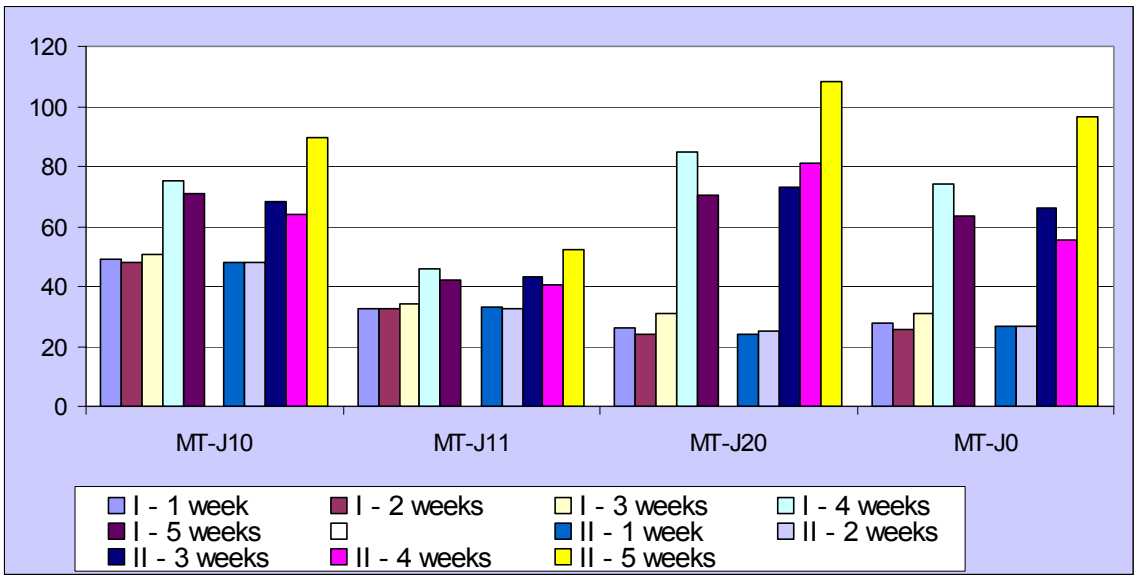

Fig. 1. Mean value of the signal maxima of MOX sensors of the beef samples with the pH-value of 5.5 (I) and of 5.9 (II) during ageing.

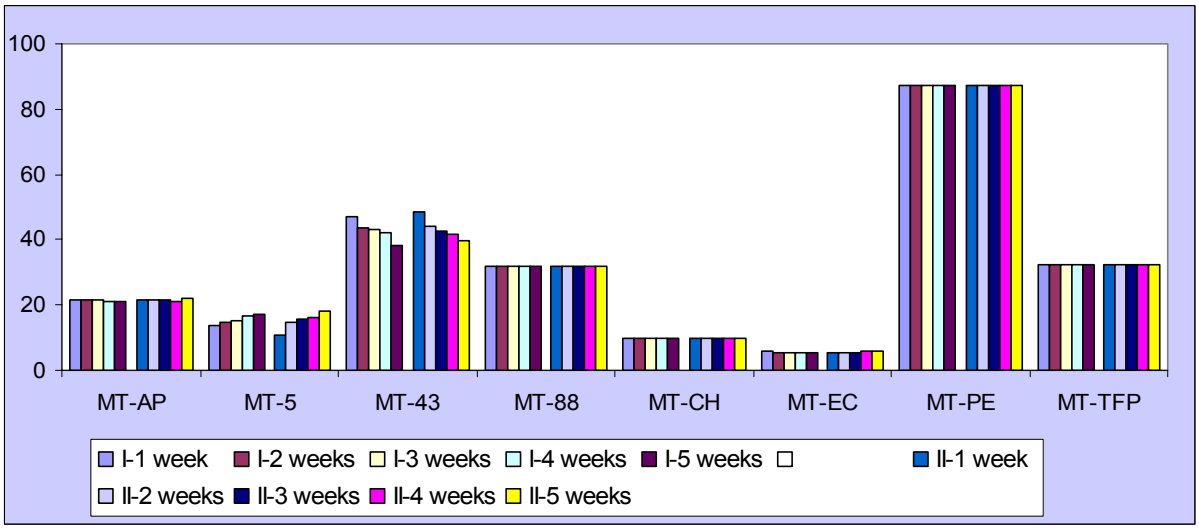

Fig. 2. Mean value of the signal maxima of QMB sensors of the beef samples with the pH-value of 5.5 (I) and of 5.9 (II) during ageing.

A smaller decrease in the signal intensity of storage of 1 to 5 weeks was determined for the sensor MT-43 specified for medium polar hydrocarbons and an increase in the signal intensity was determined for the sensor MT-5 reacting on nonpolar hydrocarbons. On the basis of the signal intensity, those sensors reacting on the differences in the meat flavour were selected and taken up for further evaluation.

In Figure 3 you can see the principal component analysis of the beef samples with different initial pH-values for the ageing process. The evaluation only contains the sensor signals reacting on differences in the meat flavour. Here it proved to show that the meat samples with a different storage period and a different pH-value can be separated by means of their sensor signals.
The differently evaluated samples in the multidimensional space are separated in a way that the differences of the sensorial testing can be found in the distances. You can see smaller distances between the clusters of the samples whose flavour has become a similar evaluation. The “good” and “bad” samples with respect to tenderness and flavour are placed in one area of the illustration. Larger differences of the gas composition

were caused by the ageing of the beef samples with the pH-value of 5.9 compared to the samples with the pH-value of 5.5.

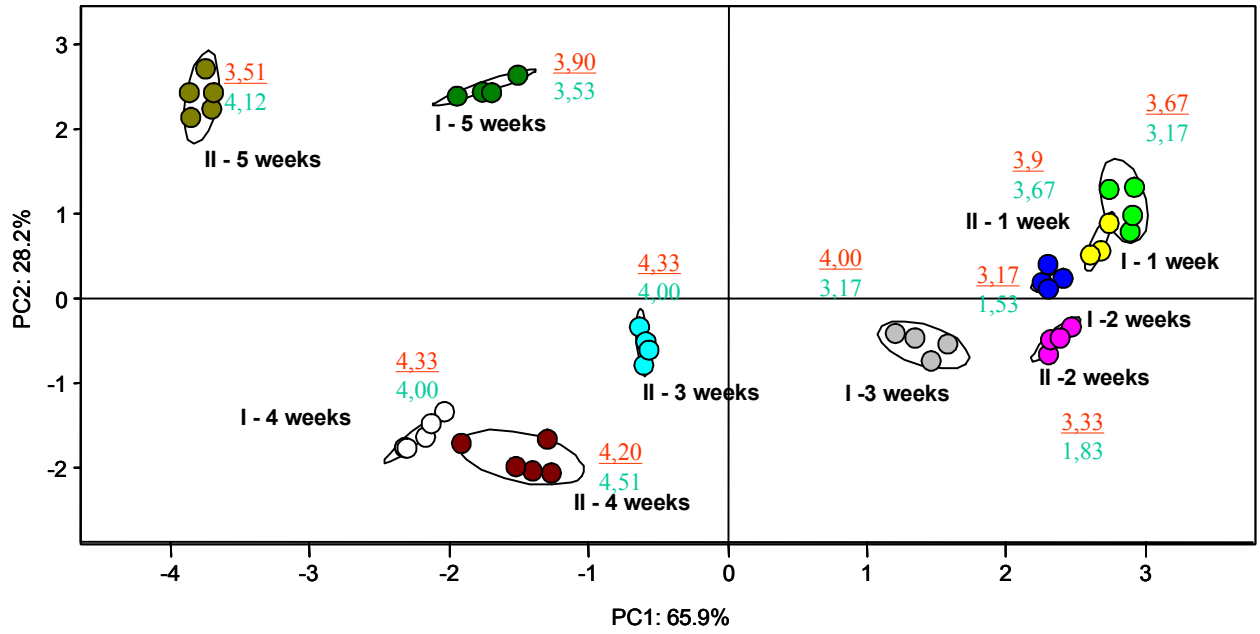

Fig. 3. Principal component analysis of the beef samples with a different pH-value during ageing (I –pH 5.5; II – pH 5.9). 3.67/3.17 – sensorial established values: Flavour/tenderness on the basis of a 6-point scheme.

The principal components analysis showed that the meat samples with a different storage period and a different pH value can be separated by means of their Headspace gas composition.

Conclusions

Due to this information, it would be possible to replace sensors with low distinctiveness by sensors of another selectivity being more relevant for the problem of separation (meat ageing), thus increasing the distinctiveness of the Cassensor-Arrays.

The available results show that the chosen sensor arrangement of 4 MOX sensors and 2 QMB sensors has been able to recognize the ageing condition of beef samples with a different pH-value corresponding to the sensorial evaluation.

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SENSORIAL AND RHEOLOGICAL PROPERTIES OF "PAINHO DE PORTALEGRE". THE INFLUENCE OF MEAT AND FAT SIZE PORTIONS AND NACL CONCENTRATION.

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Background

Traditional products of processed meat products have a positive impact on the sustainability of regional economy. Their consumption, mainly in those using raw materials from Alentejano pig breed, present some variability due to changes occurred on processing practices. Among these, meat and fat mincing size and the level of salt (NaCl) addition could fulfil a great influence on sensory and textural properties of the final product.

Objectives

The purpose of this work is to evaluate the rheological and sensorial properties occurred in "Painho de Portalegre" using low levels of salt in the formulation (0,5% and 1% NaCl) and two different minced meat called small portions and large portions.

Material and methods

"Painho de Portalegre" is a regional and traditional portuguese smoked sausage (cylindrical shape; length 30cm; diameter 25 to 40 mm) made from meat and fat obtained from Alentejano pig, a Portuguese breed.
The small portions refers to parallelepiped portions 1,7 X 1,7cm and large portions to 5,5 X 2,5cm. Two different concentrations of salt, 0,5% and 1% NaCl, were used in the formulation. Three samples of final product were used for each one of the 4 modalities (small portions with 0,5% NaCl, in the formulation; small portions with 1% NaCl, in the formulation; large portions with 0,5% NaCl; large portions with 1% NaCl).
The sensorial evaluation was a descriptive and quantitative analysis, considering a scale from 1 to 100, and the number of panellists was 12. The attributes considered were Colour Intensity, Aroma Intensity, Taste Intensity, Tenderness, Fibrouseness, Juiciness, Undesired Taste, Salt and General Appreciation.
A Texture Profile Analyze (TPA) with a compress platen and a cutting test with a blade knife was performed using a Stable Micro System TA-Hdi.in order to define the texture and compare it with the sensorial evaluation. The samples for the first test were cylindrical with 3,5cm of diameter and 3,5cm of height and were compressed twice to 10% of the initial height. For the cutting test the samples were slices with 4 mm of height and the cut was until reach 87,5% of the sample and the maximum force was measured.
Results were analysed trough an ANOVA-MANOVA considering 2 factors (salt concentration and minced) and their interactions, using the STATISTICA software. A Tukey test for comparation of means was done too.

Results and Discussion

None significant differences were found in the sensorial evaluation results (Table 1). However can be noticed that samples obtained from "Small Portions" exhibit higher values of aroma intensity and fibrouseness lower than those of "Large Portions" (Table 2).
Samples obtained with 1% NaCl exhibit higher values of tenderness (confirmed by rheological results), juiciness and taste intensity. This high classification of the two last attributes is possibly due to the salt effect on the salivation and increasing the flavour of food. Samples with 0,5% NaCl showed higher intensity of colour than those with 1% NaCl
Panellists only distinguish salt intensity for the samples produced with "Small Portions" (56 points for sausages with 0,5% NaCl and 64 points for those with 1% NaCl).
The general appreciation revealed a clear preference by the sausages produced with "Small Portions" and 0,5% NaCl.
Coutron-Gambotti *et al*. (1999) studied the effect of low concentration of salt under lipid composition and the sensorial attributes in dry ham and they observed decreasing of auto oxidation processes and a better aroma and flavour.
The analysis of the rhelogical results revealed that cohesiveness showed significant differences ($p<0,01$) for factor Minced (average value 0,74 for "Small Portions" and 0,68 for "Large Portions"). On the other hand all the other parameters didn't exhibit significant differences. Results of springiness, chewiness and cohesiveness were superior for

"Small Portions" than those of "Large Portions" due to a better binding of the meat and fat portions because after prepared they had higher contact area among them, so the extraction of soluble protein is higher too. This is corroborated by high values of cutting test found in the sausages obtained with "Small Portions" and this parameter was significant for the factor Minced ($p<0,05$).

Conclusions

A general approach of the results of Sensorial Evaluation conclude that sausages obtained through "Small Portions" exhibit a better classification than those obtained from "Large Portions". Panellists also noticed a high concentration of salt in the products from "Small Portions" and more intensity of aroma. Although none of the sensorial parameters showed significant results from Anova.
Products with 1% of NaCl (in the formulation) were classified with higher values of tenderness, juiciness and taste but lower colour intensity when compared with those with 0,5% of salt (in the formulation).
It is possible to produce sausages of high level of quality using the usual technology but with lower concentration of salt.

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Table 1 – Anova analysis for sensorial evaluation considering two factors Minced **(M)** and Na Cl concentration **(S)** and their interaction

	Minced (M)			**NaCl concentration (S)**			**Interaction (M x S)**		
	F	**p**	**Significant level**	**F**	**p**	**Significant level**	**F**	**p**	**Significant level**
Colour Intensity	0,886	0,3605	NS	3,950	0,0643	NS	0,175	0,6812	NS
Aroma Intensity	0,341	0,5675	NS	0,123	0,7307	NS	0,085	0,7741	NS
Tenderness	0,287	0,5996	NS	1,814	0,1968	NS	0,045	0,8356	NS
Fibrouseness	0,675	0,4233	NS	0,102	0,7538	NS	0,224	0,6421	NS
Juiciness	0,548	0,4704	NS	0,985	0,3367	NS	0,096	0,7609	NS
Taste Intensity	0,076	0,7863	NS	1,217	0,2863	NS	0,149	0,7045	NS
Undesired Taste	0,051	0,8243	NS	0,354	0,5602	NS	0,0004	0,9839	NS
Salt Intensity	1,043	0,3223	NS	0,718	0,4092	NS	0,794	0,3861	NS
General Appreciation	0,898	0,3573	NS	0,003	0,9544	NS	0,135	0,7180	NS

Legend: NS – no significant, $p \geq 0,05$

Table 2 – Means and Standard deviation of Sensorial Evaluation considering two factors Minced **(M)**, NaCl concentration **(S)**

Minced	NaCl (%)	Colour Intensity	Aroma Intensity	Tenderness	Fibrouseness	Juiciness	Taste Intensity	Undesired Taste	Salt Intensity	General Appreciation
Small Portions	0,5	75,6 ± 9,317	69,4 ± 12,260	65,0 ± 16,583	31,4 ± 22,109	66,75 ± 9,708	67,6 ± 10,550	5,2 ± 8,438	56,0 ± 8,944	71,2 ± 11,189
	1	61,8 ± 18,199	65,0 ± 8,775	74,6 ± 14,519	29,8 ± 19,880	70,0 ± 10,000	72,2 ± 5,310	8,0 ± 13,038	64,0 ± 11,402	69,0 ± 6,519
Large Portions	0,5	78,6 ± 11,824	63,4 ± 15,550	69,6 ± 12,280	35,0 ± 19,365	68,8 ± 8,786	65,2 ± 5,805	4,0 ± 8,944	55,4 ± 9,788	64,4 ± 9,317
	1	69,6 ± 10,015	63,0 ± 21,679	76,6 ± 11,082	43,2 ± 29,685	75,0 ± 12,329	70,6 ± 9,476	7,0 ± 12,410	55,2 ± 10,849	66,0 ± 16,733

Table 3 – Anova analysis for rheological evaluation considering two factors Minced **(M)** and NaCl concentration **(S)** and their interaction

	Minced (M)			NaCl concentration (S)			Interaction (MxS)		
	F	p	Significant level	F	p	Significant level	F	p	Significant level
Hardness (N)	0,075	0,7867	NS	0,112	0,7413	NS	0,035	0,8541	NS
Cohesiveness	9,368	0,0062	**	0,002	0,9633	NS	0,031	0,8616	NS
Springiness	2,487	0,1305	NS	0,159	0,6941	NS	0,211	0,6512	NS
Gumminess (N)	0,008	0,9276	NS	0,084	0,7744	NS	0,015	0,9023	NS
Chewinness (N)	0,178	0,6776	NS	0,015	0,9048	NS	0,0001	0,9932	NS
Cutting Test (N)	7,131	0,0147	*	0,023	0,8801	NS	0,042	0,8392	NS

Legend: NS – no significant, p≥0,05; * - significant, p<0,05; ** - significant, p<0,01

Table 4–Means and Standard deviation for rheological evaluation considering two factors Minced **(M)**, NaCl concentration **(S)**

Minced	NaCl (%)	Hardness (N)	Cohesiveness	Springiness	Gumminess (N)	Chewinness (N)	Cutting Test (N)
Small Portions	0,5	1005,997 ± 825,559	0,744 ± 0,037 a	0,842 ± 0,068	761,274 ± 650,757	648,664 ± 571,598	2321,676 ± 524,609 a
	1	969,968 ± 345,202	0,740 ± 0,049 a	0,840 ± 0,091	730,524 ± 294,025	628,042 ± 282,617	2375,954 ± 679,956[a]
Large Portions	0,5	1117,761 ± 624,355	0,682 ± 0,040b	0,761 ± 0,096	767,245 ± 420,917	580,193 ± 293,899	3608,670 ± 1729,545b
	1	991,331 ± 471,762	0,684 ± 0,060b	0,795 ± 0,127	690,489 ± 367,084	562,301 ± 337,990	3532,009 ± 1271,392b

Acknowledgements
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LIPID OXIDATION DURING REFRIGERATED STORAGE OF LIVER PÂTÉS FROM EXTENSIVELY REARED IBERIAN PIGS AND INTENSIVELY REARED WHITE PIGS

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Background

Iberian pigs have been traditionally reared extensively in evergreen-oak forests named 'dehesas' in which animals are fed on natural resources (grass and acorns). On the other hand, pigs from industrial genotypes are selected for high growth rates and traditionally reared intensively, under controlled conditions and fed with concentrated diets, in order to maximize benefits in the shortest period of time (Sundrum, 2001).
Oxidation of lipids is the major factor reducing quality and acceptability of meat and fat products (Morrissey et al., 1998). Lipid oxidation is a complex process whereby polyunsaturated fatty acids (PUFA) are degraded via formation of free radicals, causing flavour, texture, colour and nutritional deterioration of foodstuffs (Morrissey et al., 1998).
Liver pâté is a traditional product with excellent nutritional and sensory properties (Echarte et al., 2004). Ingredients (mainly liver, fat and meat) are finely minced and cooked during pâté manufacture that could favour the development of lipid oxidation. Thus, liver pâtés exhibit high amounts of fat and iron, and therefore, oxidative deterioration of liver pâtés during refrigeration is expected. The differences between pâtés from Iberian and white pigs in terms of their fatty acid composition and antioxidative status (Estévez et al., 2004) are expected to influence on their oxidative deterioration during refrigerated storage.

Objectives

The aim of the present work was to study the lipid oxidative changes of liver pâtés from Iberian and white pigs during refrigerated storage as assessed by PUFA degradation and generation of thiobarbituric acid reactive substances (TBA-RS) and hexanal.

Material and methods

Animals: Iberian pigs were free-range reared and fed on natural resources (grass and acorns) following the traditional livestock farming for Iberian pigs. The animals were slaughtered at ~150 kg live weight and at the age of 12 months. White pigs (Large White x Landrace) were intensively reared under controlled conditions in a typical industrial livestock farm. The animals were fed on a concentrate feed and slaughtered at 85 kg live weight and at the age of 7 months.
Samples, manufacture of pâté and refrigerated storage: After slaughter, back fat, muscle *Quadriceps femoris*, and liver were removed from the carcasses, vacuum packaged and stored at -80ºC until the day of the manufacture of the pâté. For this, ingredients were as follows per 100g of elaborated product: 28g liver, 40g adipose tissue, 5g muscle, 23g distilled water, 2g sodium caseinate, 2g sodium chloride. Sodium di- and tri-phosphates (0.3%) sodium ascorbate (0.05%) and sodium nitrite (0.03%) (ANVISA, Madrid, Spain) were also added. The protocol followed for the manufacture of liver pâtés was described elsewhere (Estévez et al., 2004). Liver pâtés were packed in glass containers prior to thermal treatment (80ºC/30min.). Liver pâtés were stored in the darkness at 4ºC during 90 days since the day of the manufacture (day 0). Liver pâtés were analysed at days 0, 30, 60 and 90 for PUFA content, TBA-RS and hexanal. After being accomplished each of the refrigeration stages, liver pâtés were stored at -80ºC until analytical experiments.
Chemical Analysis: Moisture, total protein, total fat, and ash were determined using official methods (AOAC, 2000). Fatty acid methyl esters (FAME) were prepared following the method of López-Bote et al. (1997). FAME were analysed using a Hewlett Packard, gas chromatograph, equipped with a flame ionisation detector (FID) as described Estévez et al. (2004). Identification of FAME was based on retention times of reference compounds (Sigma). Proportions of saturated, monounsaturated and polyunsaturated fatty acids (SFA, MUFA and PUFA, respectively) were calculated as percentages of total fatty acids analysed. The quantification of PUFA (sum of C18:2, C18:3 and C20:4) was carried out by using C13 as internal standard. Results are expressed as g PUFA $100g^{-1}$ pâté. TBARS were determined using the method of Rosmini et al.,

(1996). Hexanal was determined in the headspace of liver pâtés using the solid-phase microextraction (SPME) sampling coupled to gas chromatography and mass spectrometry (GC-MS) (Estévez et al., 2003).
Data Analysis: Results of the experiments were used as variables and analysed by using a Student's t-test for independent variables (SPSS, 1997) in order to compare pâtés from Iberian and white pigs. The effect of refrigerated storage on liver pâtés was assessed by using an Analysis of Variance (ANOVA) from SPSS software. Statistical significance was considered as follows: $p>0.05$ (ns), $p<0.05$ (*), $p<0.01$ (**), $p<0.001$ (***).

Results and Discussion

Liver pâtés from Iberian and white pigs showed no differences for moisture, fat, protein or ash contents (Table 1). However, compared to pâtés from white pigs, pâtés from Iberian pigs presented a larger proportion of MUFA and smaller of SFA and PUFA. Liver pâtés reflected the fatty acid composition of the raw material (fat, liver and meat) used for their manufacture (Estévez et al., 2004). Muscles, back-fat and liver tissues from Iberian pigs present large amounts of oleic acid and MUFA as a result of the intake of acorns during the last stage of their fattening period that has been linked to several quality traits (López-Bote et al., 1997).
The amount of PUFA gradually decreased in both types of pâté during refrigerated storage as a likely consequence of the development of oxidative reactions. The decrease rate of PUFA content as measured by the slopes of the calculated trend lines (Figure 1) revealed that the loss of PUFA was more intense in pâtés from white pigs than in the Iberian counterparts. After 90 days of refrigerated storage, pâtés from white pigs lose 2.3 g PUFA/100g pâté as an average while those from Iberian pigs lose a significantly ($p<0.05$) smaller amount (1.5 g PUFA/100g pâté). The oxidative degradation of PUFA led to a gradually increase in the amount of TBA-RS and hexanal between day 0 and day 90 for liver pâtés from Iberian and white pigs (Figure 2). Pâtés from white pigs presented significantly ($p<0.05$) higher TBA-RS numbers and hexanal contents than pâtés from Iberian pigs at day 0 and day 90. These results are consistent with those obtained from the PUFA degradation and agree with those obtained in previous works devoted to the study of the oxidation stability of raw and cooked meats from Iberian and white pigs (Estévez et al., 2003; Estévez et al., 2004) and could be partly explained by the equilibrium between prooxidant and antioxidant factors in the pâtés. Pâtés from white pigs presented a higher proportion of PUFA and lower of MUFA than pâtés from Iberian pigs that makes the former more prone to oxidation than the latter. Moreover, in a previous work (Estévez et al., 2004) we reported a significantly higher amount of vitamin E in the raw material used for the manufacture of pâtés (fat, liver and muscles) from Iberian pigs when compared to those from white pigs. The relationship between the nutritional background (pasture- and mixed diet finishing) and the fatty acid profile and oxidation stability of liver, pork and on them based products is profusely documented (Cava et al., 2000; Nilzén et al., 2001). The intake of pasture by animals increases in their tissues the level of vitamin E, enhancing their oxidation stability (Cava et al., 2000; Nilzén et al., 2001).

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Table 1. Chemical composition and fatty acid profile of liver pâtés from extensively reared Iberian pigs and intensively reared white pigs.

	Iberian	white	P[2]
Moisture[1]	48.42±1.37	50.51±0.62	ns
Protein[1]	10.34±0.24	10.04±0.70	ns
Ash[1]	2.69±0.09	2.78±0.21	ns
Fat[1]	33.37±1.81	31.82±0.57	ns
Fatty acids[3]			
SFA	32.87±0.09	37.98±0.12	***
MUFA	57.52±0.06	47.58±0.10	***
PUFA	9.63±0.11	14.40±0.25	***

[1]g/100g pâté; [2]Statistical significance; [3]Percentage of total methyl esters analysed
SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids
ns: non significant; *: p<0.05; **: p<0.01; ***: p<0.001

Figure 1. Trend lines of the evolution of PUFA amounts (g/100g pâté) during refrigerated storage of liver pâtés from extensively reared Iberian pigs and intensively reared white pigs.

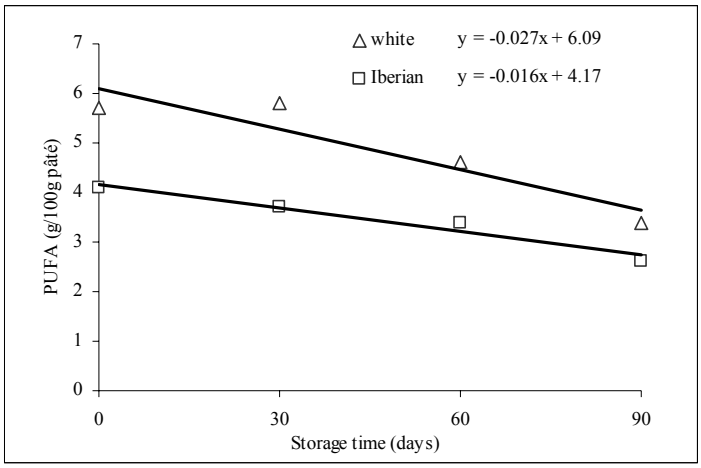
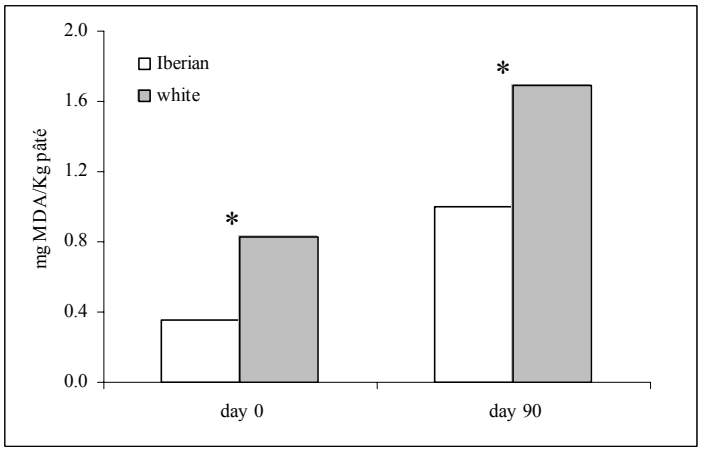
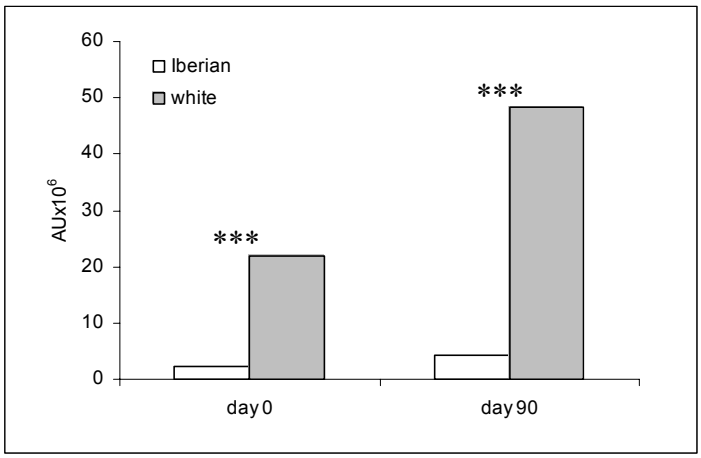


Figure 2. TBA-RS in liver pâtés from extensively reared Iberian pigs and intensively reared white pigs before (day 0) and after 90 days of refrigerated storage (day 90).



ns: non significant; *: p<0.05; **: p<0.01; ***: p<0.001

Figure 3. Hexanal content in liver pâtés from extensively reared Iberian pigs and intensively reared white pigs before (day 0) and after 90 days of refrigerated storage (day 90).



ns: non significant; *: p<0.05; **: p<0.01; ***: p<0.001

EFFECT OF THE ADDITION OF SAGE AND ROSEMARY EXTRACTS ON THE OXIDATIVE STABILITY OF DIFFERENT TYPES OF LIVER PÂTÉS

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Background

Liver pâté is an emulsion-like product that exhibits high contents of fat and iron that would make of it a product with high oxidative instability (Russell et al., 2003; Estévez et al., 2004). The pâté matrix is relatively poor in natural antioxidants, which justifies the addition of exogenous antioxidants (Madhavi et al., 1996) in order to inhibit the development of the aforementioned oxidative reactions. Synthetic phenolic antioxidants, such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and propyl, octyl and dodecyl gallates (PG, OG, DG), are easily available and largely used in different food products (Pinho et al., 2000). Using such synthetic compounds has been linked to health risks generally referred to carcinogenic potential (Clayson et al., 1986). Consequently, many scientific efforts have been led to select different natural antioxidant extracts and prove their antioxidant activities as alternatives to synthetic antioxidants. Sage (*Salvia officinalis*) and rosemary (*Rosmarinus officianalis*) are popular *Labiatae* herbs with a verified potent antioxidant activity in meat and fat products (McCarthy et al., 2001; Yu et al., 2002). On the other hand, the compositional characteristics of the raw material influence on the quality characteristics and oxidative stability of the manufacture product (Estévez et al., 2004). Liver pâtés from extensively reared Iberian pigs demonstrated to have a higher oxidative stability than pâtés from intensively reared white pigs as a result of the fatty acid composition and the presence of higher amounts of endogenous antioxidants such as vitamin E (Estévez et al., 2004).

Objectives

The aim of the present work were to investigate the effect of the addition of natural antioxidants (extracts of sage and rosemary) on the oxidative stability of refrigerated stored pâtés from extensively reared Iberian pigs and intensively reared white pigs.

Material and methods

Animals: Iberian pigs were free-range reared and fed on natural resources (grass and acorns) following the traditional livestock farming for Iberian pigs. White pigs (Large White x Landrace) were intensively reared under controlled conditions in a typical industrial livestock farm a fed on a concentrate feed.
Samples, manufacture of pâté and refrigeration: After slaughter, back fat, muscle *quadriceps femoris*, and liver were removed from the carcasses, vacuum packaged and stored at -80ºC until the day of the manufacture of the pâté. For this, ingredients were as follows per 100g of elaborated product: 28g liver, 40g adipose tissue, 5g muscle, 23g distilled water, 2g sodium caseinate, 2g sodium chloride. Sodium di- and tri-phosphates (0.3%) sodium ascorbate (0.05%) and sodium nitrite (0.03%) (ANVISA, Madrid, Spain) were also added. Depending on the origin of the raw material (from 'Iberian' or 'white' pigs) and the addition of different antioxidants ('rosemary' and 'sage' extracts, 0.1%) different types of pâtés were elaborated. A 'control' batch without added extract was also considered. The protocol followed for the manuacture of liver pâtés was described elsewhere (Estévez et al., 2004). Liver pâtés were packed in glass containers prior to thermal treatment (80ºC/30min.). After manufacture, liver pâtés were stored in the darkness at 4ºC during 90 days in order to allow the development of oxidative reactions. Liver pâtés were analysed at days 0 and 90 for the amount of reactive substances to the tiobarbituric acid (TBA-RS), increase of carbonyls from protein oxidation and hexanal.
ChemicalAnalysis: TBA-RS were determined using the method developed by Rosmini et al., (1996) for liver pâtés. Protein oxidation as measured by the total carbonyl content was assessed following the method described by Oliver et al., (1987). Hexanal was determined in the headspace (HS) of liver pâtés using the solid-phase microextraction (SPME) sampling coupled to gas chromatography and mass spectrometry (GC-MS) (Estévez et al., 2003).

Data Analysis: Results from the experiments were used as variables and analysed by using a Student's t-test for dependent variables (SPSS, 1997) in order evaluate significant changes between days 0 and 90. The effect of the addition of the antioxidants was assessed by using an one-way Analysis of Variance (ANOVA) from SPSS software. When statistical differences were detected, data were analysed using Tukey's tests. Statistical significance was set at $p<0.05$.

Results and Discussion

The development of oxidative reactions during refrigerated storage led to a gradual increase in the amount of residual components in liver pâtés such as those generated from lipids and proteins. TBA-RS significantly increased ($p<0.05$) in all groups during refrigerated storage (Figures 1A, 2A). Similarly, the amount of carbonyls from protein oxidation had significantly increased ($p<0.05$) after 90 days of refrigerated storage in control and treated pâtés (Figures 1B, 2B). Due to significant correlations found between some particular lipid-derived volatiles and other lipid oxidation products such as TBA-RS (Shahidi & Pegg, 1994), hexanal has been considered as a fairly good indicator of lipid oxidation. During refrigerated storage, the hexanal significantly ($p<0.05$) increased in the pâtés HS (Figures 3A, 3B), agreeing with the results obtained from the oxidation of lipids and proteins. In general, the addition of sage and rosemary essential oils had a significant effect on the oxidative stability of liver pâtés but this effect was different depending on whether they were added on 'Iberian' or 'white' pâtés. In agreement with previous research on several meats and meat products (McArthy et al., 2001; Yu et al., 2002), the addition of sage and rosemary essential oils had an antioxidant effect on pâtés from Iberian pigs as long as smaller amounts TBA-RS, carbonyls from protein oxidation and hexanal were detected in treated pâtés when compared to the 'control' counterparts. In contrast, the addition of sage and rosemary in pâtés from white pigs had an opposite behaviour, significantly increasing ($p<0.05$) the generation of lipid and protein oxidation products, while no effect was detected for the generation of hexanal. Results from the present work suggest that the activity of sage and rosemary essential oils is dependent on the compositional characteristics of the food matrix. Food systems, and specifically liver pâtés, are very complex in the number and the type of chemicals in the mixture, and a combination of these compounds may behave differently from the individual components. In this sense, Wong et al. (1995) and Fang & Wada (1993) reported possible interactions between phenolic compounds from sage and rosemary essential oils and vitamin E, resulting in different activities depending on the individual amounts of these substances in the food system. Significant differences ($p<0.05$) were found between Iberian and white pigs regarding vitamin E content in muscles (6.18 vs 1.94 mg/kg muscle), livers (7.93 vs 3.49 mg/kg liver) and adipose tissues (19.67 vs 1.21 mg/kg adipose tissue) used for the manufacture of liver pâtés (Estévez et al., 2004) as a likely consequence of the intake of pasture during fattening. The presence of a certain amount of an endogenous antioxidant (vitamin E) in the raw material and manufacture product might influence on the activity of exogenous active extracts, leading to antioxidant or pro-oxidant effects.

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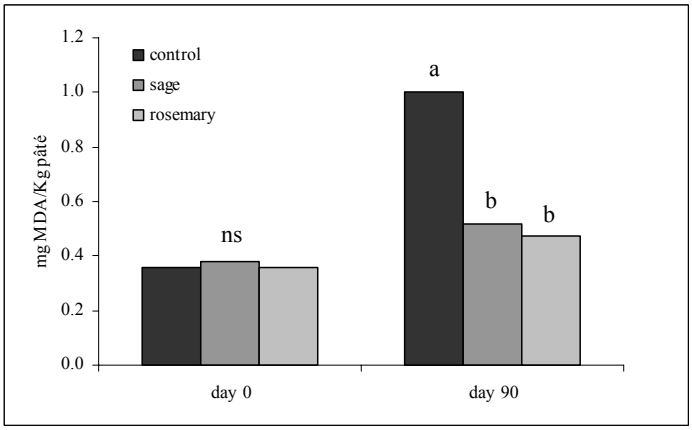
Figure 1. TBA-RS numbers (A), carbonyls from protein oxidation (B) and hexanal content (C) in pâtés from **IBERIAN PIGS** as affected by the addition of sage and rosemary essential oils

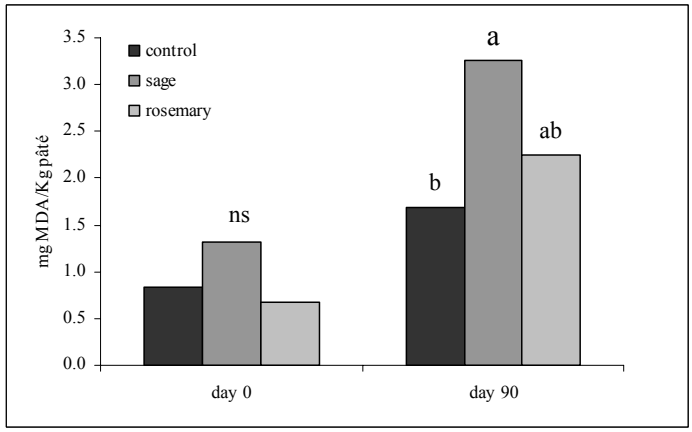
ns: non significant; a,b: different letters denote differences betweeen groups withinh a day

Figure 2. TBA-RS numbers (A), carbonyls from protein oxidation (B) and hexanal content (C) in pâtés from **WHITE PIGS** as affected by the addition of sage and rosemary essential oils

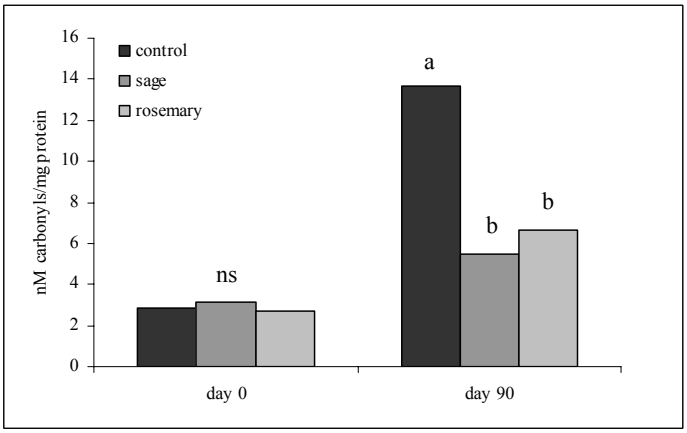
ns: non significant; a,b: different letters denote differences betweeen groups withinh a day

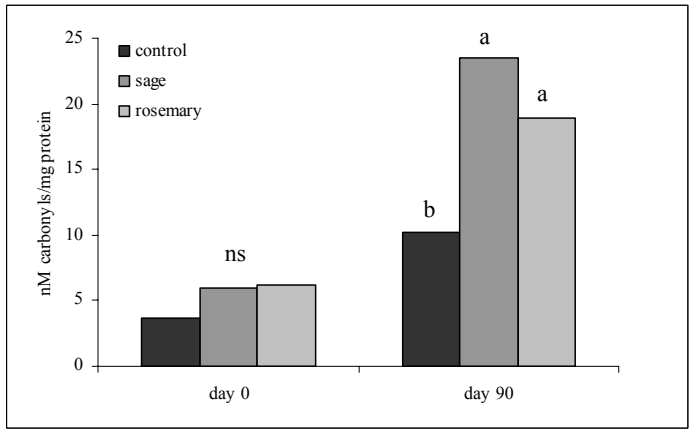
(A) TBA-RS NUMBERS



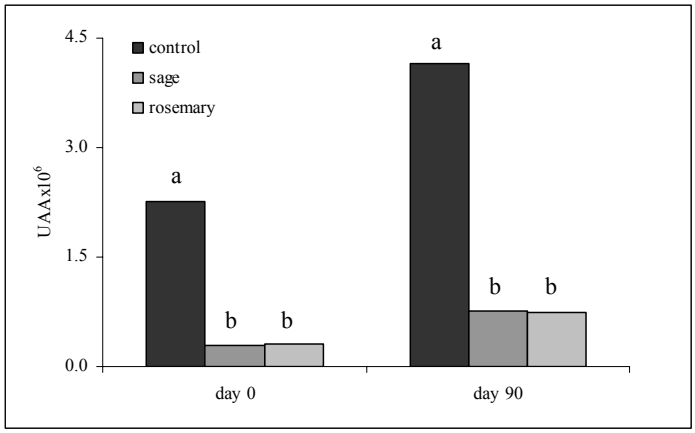


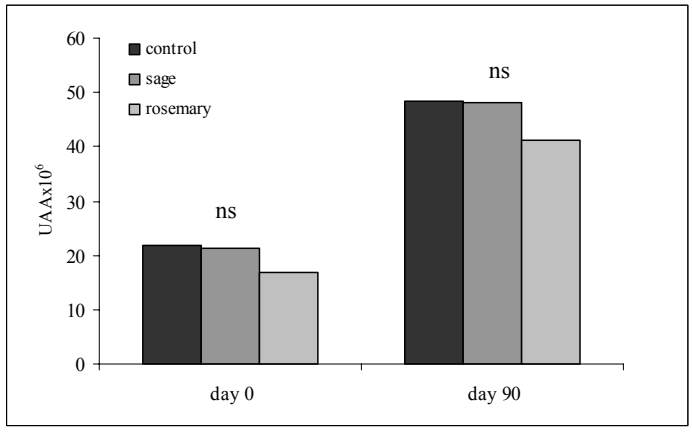
(B) CARBONYLS





(C) HEXANAL





ANALYSIS OF UNIQUE FLAVOR OF CHINESE TRADITIONAL XUANWEI HAM

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Background

The cured ham was initially intended to solve the problem of storing fresh meat. But now with the wide use of the refrigeration, the major attention has been shifted to its unique flavor. A typical flavor can provide a pleasant appreciation and wide acceptance for customers. For example, Iberian Ham has become one of the most famous meat products of high quality in Europe. Xuanwei Ham is a famous special product in Yunnan Province of China. At present, studies on the characteristics of Xuanwei Ham are not enough.

Objectives

This research was conducted to analyze the impact of the climate character in Xuanwei area, raw materials and traditional techniques on the quality of traditional Xuanwei ham.

Materials and Methods

Ham Process: Wujin Pigs were slaughtered when they grew to 8~12 month old, 90~100kg. Hams were made in traditional procedures. Green hams were held for 12~24h at 5~10℃ after slaughter. The hams were thoroughly rubbed with local salt and placed in piles with alternate beds of hams and salted for 30 days at 5~10 ℃ and 65%~85% relative humidity. The salt was added on the proportion of 7% of green legs and was rubbed two or three times at a 7 day interval. The salting started from late October and ended early February in the following year. After washing to remove the excess salt and dirt on the surface of the ham, they were hung with straw strings and dried in the sunlight for 3~4 days. The hams were hung on the strings to ripen for 8~12 months in a ventilated chamber (temperature ranging from 10~20℃ and relative humidity 60%~80%).

Sampling. The *Biceps femoris* were taken from 10 hams of 12-month maturation and the physicochemical and the volatile compounds were analyzed and identified immediately.

Chemical Analyses. Sodium chloride was determined by potentiometric titration with $AgNo_3$ in an autotitrator, and results are given as a percentage of NaCl (w/w). *Water content* was determined after drying of a 3.00 g homogenized sample at 105℃ for 18 h and subsequent cooling in a desiccator. Results are given as a percentage of water (w/w). *Water activity* (a_w) was determined at 25℃ by the graphic interpolation method using saturated salt solutions as standards. *Free amino acids*: Samples of 1 g of ham was homogenized with 25 ml of sulphosalicylic acid (8%) in a 50ml centrifugal tube and the tube was set in ice. The homogenate was centrifuged in a refrigerated centrifuge at 8000 rpm for 13 min. The supernatant was filtered through Whatman No. 54 filter paper. Some amino acids were identified and quantified were measured by L-8500 Amino Acid Analytical Apparatus. *Volatile compounds analysis*: The volatile compounds were extracted by employing a simultaneous distillation-extraction method (SDE), and were separated and identified by Agilent 6890 Series gas chromatographer (GC) and Agilent 5973 Network Mass Selective Detector (MS).

Result and Discussion

1. Physicochemical analysis of traditional Xuanwei ham

The production yield of traditional Xuanwei ham maintained about 69.30%~73.60% (Table 1). The proportion of muscle to fat varied according to the composition of raw hams. The raw hams usually come from pigs with more fat and thus, the finished ham is rich in fat.

The traditional Xuanwei ham is rich in salt, which has an influence on consumers' appreciation and acceptance. Compared with other dry cured ham products, there were almost no differences concerning water content and a_w in the *M. biceps femoris* and subcutaneous fat (Table 2).

The free amino acids in Xuanwei ham are rich (Table 3). The most abundant is Glutamic acid，followed by Alanine and Leucine. The total free amino acids are 9040.45mg·100g^{-1} *M. biceps femoris* dry matter, similar to the results obtained from Iberian ham[1, 2]. Iberian ham was processed with long ripening and drying periods (18~24 months) and Xuanwei ham only had shorter production periods (8 ~ 12 months). This illustrated that Xuanwei ham has characteristics of quicker ripening process, higher activity of proteolytic enzyme, more free amino acids formed than those of Iberian Ham. These characteristics may well be related to the climate character, under which Xuanwei hams were dried and ripened.

2. Separation, identification and content evaluation of volatile compounds in Xuanwei ham

Separating and identifying results of the volatile compounds in the *Biceps femoris* of traditional Xuanwei ham were given in table 4.

Results showed that 75 volatile components were tentatively identified in the volatile fraction, including 15 hydrocarbons, 9 alcohols, 22 aldehydes, 6 ketones, 3 acids, 7 esters and 13 others. In the recent years, studies on volatile aroma compounds about Iberian ham, Serrano ham, Parma ham and Jinhua ham have been carried out by some experts[3, 4, 5, 6]. Some of the volatile compounds separated and identified from traditional Xuanwei ham had some similarities to those results reported of those dry cured ham products, but there were some differences. For example, in our samples, less species of ketones, esters and low molecular weight compounds were identified, but more branched compounds were obtained. Short chain hydrocarbon compounds less than C_{12} were not found. Such differences were correlated with experimental methods, sampling time and materials. Therefore, as regards the typical flavor properties and the exact quantity of the volatile compounds, it is hard to identify them and needs more careful future study.

3. The traditional Xuanwei ham is the work of nature and the special terrain

Xuanwei City is located in the north-east of Yunnan Province in China, east longitude 103º35′ 30″ ～ 104º 40′ 50″, north latitude 25º53′ 30″ ～ 26º44′ 50″. It is located in the transitional draping belt of Yuannan Plateau and Guizhou Plateau, its elevation is 2,868 to 920 meters. The winter begins from November 6th and ends on March 5th of the next year, and the spring begins from March 6th and the autumn ends on November 15th. There is no summer. The main weather reports of Xuanwei town are seen in the table 5. The unique weather creates an appropriate natural climate environment of curing, drying and ripening Xuanwei ham. The hams cured in winter were dried and ripened in spring and autumn. Therefore, Xuanwei City is a natural workshop of dry-cured hams. The natural climate character is the key factor of forming high quality.

4. The special pig specie and feeding way are the quality guarantee of the traditional Xuanwei ham

Wujin pig is a typical local specie. It has higher ability of digesting crude feedstuff. It not only adapts to the climate of high and cold mountain area, but also the warm and hot river valley area. Its body fat is high, it grows slowly, and the quality of muscle is good. In the early growth stage, the greens, straw chaff and rice bran were primary feedstuff. When the weight reaches to 40～50kg, proportion of corn flour and potato were increased in primary feed. The common green stuff are the straw chaff of horsebean, pease, buckwheat and sweet potato, the wild feedstuff are sweet potato vine, potato, carrot, pumpkin, chayote, cress, and so on. According to the feeding way of the local farmers, at the beginning, the piglet is 6.5kg, after feeding for 323 days, its weight can reach to 83.5kg. The daily average weight increase is 239g. It needs about 2.34kg foodstuff and 24.25kg green stuff to increase 1kg.

Wujin pigs' intramuscular fat content is abundant(14.37±3.82%), the flesh is tender, and cooking yield is high (71.64±5.32%). The water content in buttocks muscle is 67.3%. It is an ideal material for producing the high qualtity Xuanwei ham. The aroma of ham made of Wujin pig is good than that of others. The reasons require further investigation.

Conclusion

The ratio of finished product of traditional Xuanwei ham is 69.30%～73.60%, and that the water content, salt content, a_w of *M. Biceps femoris* were 47.30%~50.50%, 9.10%～11.20%, 0.83～0.85 respectively, it has a good storage stability. The ripening ham muscle was rich in free amino acid, the free amino acid content of *M. Biceps femoris* dry matter amounted to 9040.45mg/100g, similar to that of Iberian ham. Because of the different ripening time of two finished products, the ripening process of Xuanwei ham is quicker than that of Iberia. 75 volatile compounds are tentatively identified from the extracts in the *Biceps femoris* muscle. There are 15 hydrocarbons, 9 alcohols, 22 aldehydes, 6 ketones, 3 acids, 7 esters and 13 others. The technology of Xuanwei ham showed that the unique trait of Xuanwei ham resulted from the typical climate character in Xuanwei area, the Wujin pig, and the refined traditional processing technology.

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Table 1 The ratio and composition of end products of traditional Xuanwei ham

Index	End product ratio %	Skin %	Bone %	Lean %	Fat %
Scope	69.30~73.60	7.19~7.61	13.53~13.79	52.59~56.41	22.62~26.50

Table 2. Physicochemical index of traditional Xuanwei ham

Samples	Moisture /%	a_w/ %	NaCl /%
M. Biceps femoris	47.30~50.50	0.83~0.85	9.10~11.20
M. Subcutaneous fat	2.90~3.30	0.75~0.80	0.70~0.85

Table 3. Free amino acid content of the *M. Biceps femoris* dry matter in Xuanwei ham

Amino acids	Thr	Ser	Glu	Gly	Ala	Cys	Val	Met
$Mg \cdot 100g^{-1}$	504. 81	487.13	1274.10	390.84	882.28	0.00	565.98	279.11
Amino acids	Ile	Leu	Try	Phe	Lys	His	Arg	Pro
$Mg \cdot 100g^{-1}$	456.90	869.28	316.56	469.82	1130.67	271.40	630.11	511.51

Table 5. Xuanwei City 2002 monthly average Temperature, Relative Humidity, Rainfall and the Hours of sunlight

Month	1	2	3	4	5	6	7	8	9	10	11	12
Temperature/℃	5.5	9.9	13.0	17.3	17.2	20.2	19.2	17.8	16.4	14.0	10.6	8.0
RH /%	73	65	58	49	71	74	79	80	72	76	76	68
Rainfall /mm	21	10	19	19	141	160	111	224	106	61	9	11
sunlight /h	124	194	228	253	168	177	138	153	173	159	123	177

Table 4. The volatile components of the traditional Xuanwei ham *M. Biceps femoris* (DM)

No.	Volatile components	μg/100g	No.	Volatile components	μg/100g
	Hydrocarbons		39	2-Octenal, (E)-	62.12
1	1,3-Hexadiene, 3-ethyl-2-methyl-	14.17	40	Tetradecanal	40.33
2	(3Z,5E)-1,3,5-Undecatriene	5.45	41	Pentadecanal-	85.01
3	Tetradecane	7.63	42	Hexadecanal	5784.10
4	1-Pentadecene	18.53	43	9,17-Octadecadienal, (Z)-	13.08
5	Pentadecane	51.23	44	13-Tetradecenal	456.67
6	Pentacosane	3.27	45	9-Octadecenal, (Z)-	175.47
7	Pentadecane, 4-methyl-	4.36	46	Octadecanal	567.84
8	Pentadecane, 3-methyl-	5.45		**Ketones**	
9	Hexadecane	11.99	47	Cyclohexanone	7.63
10	Cyclopentadecane	14.17	48	2-Heptanone	35.97
11	1-Octadecene	6.54	49	Ethanone, 1-(1H-pyrrol-2-yl)-	7.63
12	Triacontane	10.90	50	2-Nonanone	18.53
13	1, 13-Tetradecadiene	10.90	51	5-Nonanone, 2,8-dimethyl-	9.81
14	Oxirane, tetradecyl-	10.90	52	2-Pentadecanone	55.58
15	Oxirane, hexadecyl-	152.59		**Acids**	
	Alcohols		53	n-Decanoic acid	9.81
16	1-Pentanol	41.42	54	n-Hexadecanoic acid	47.74
17	2-Furanmethanol	16.35	55	9,12-Octadecadienoic acid (Z,Z)	21.80
18	1-Hexanol	64.30		**Esters**	
19	Cyclohexanol	41.42	56	Formic acid, octyl ester	29.43
20	1-Heptanol	32.70	57	Decanoic acid, methyl ester	5.45
21	Phenylethyl Alcohol	7.63	58	Methyl tetradecanoate	10.90
22	3-Cyclohexene-1-methanol, .alph., .alph.	5.45	59	9-Hexadecenoic acid, methyl ester	10.90
23	E, E-2,13-Octadecadien-1-ol	68.66	60	Hexadecanoic acid, methyl ester	69.75
24	Z, E-3,13-Octadecadien-1-ol	27.25	61	8-Octadecenoic acid, methyl ester	28.34
	Aldehydes		62	9-Octadecenoic acid (Z)-, methyl ester	19.62
25	Hexanal	308.44		**Others**	
26	Furfural	13.08	63	Furan, 2-ethyl-	25.07
27	Propanal, 3-(methylthio)-	56.67	64	Pyrazine, methyl-	3.27
28	Benzaldehyde	41.42	65	Pyrazine, 2,6-dimethyl-	23.98
29	2-Thiophenecarboxaldehyde	19.62	66	Phenol, 4-ethyl-	15.26
30	Octanal	110.08	67	Furan, 2-pentyl-	37.06
31	Benzeneacetaldehyde	304.08	68	Hexanoic acid, anhydride	17.44
32	2-Octenal, (E)-	25.07	69	Phenol, 4-methyl-	17.44
33	Nonanal	152.37	70	Benzene, 1-ethenyl-4-methoxy-	3.27
34	2-Nonenal, (E)-	27.25	71	Naphthalene	5.45
35	2-Decenal, (Z)-	51.23	72	Benzothiazole	2.18
36	Decanal	8.16	73	2(3H)-Furanone, 5-butyldihydro-	8.72
37	Undecanal	5.45	74	Naphthalene, 2-methyl-	2.18
38	2,4-Decadienal	30.52	75	Naphthalene, 2,7-dimethyl-	2.18

CALCIUM CHLORIDE MARINATION OF BOVINE BRACHIOCEPHALICUS MUSCLE: EFFECT OF TUMBLING ON TEXTURAL PROPERTIES

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Background

Meat tenderness is one of the most important attributes for consumers and depends on a great number of intrinsic biological factors, as breed, age, sex, feeding, and kind of muscle, besides the ante- and postmortem conditions (Thompson, 2002). Meat ageing is an alternative to achieve more tenderness and improve the sensory and nutritional quality. Nonetheless, in Mexico and many countries in the world ageing or maturation process is not a common practice, due to higher storage cost and energy required. The tenderness obtained during meat ageing is attributed to the action of two endogenous enzymatic systems, calpains and cathepsins (Jaarseveld et al., 1997). Cathepsins are liberated in lysosomes due to the lipoprotein membrane breakdown caused by the low pH values achieved in the post-mortem muscle. Calpains are located in cells cytoplasm, and they can be activated by the calcium stored in the sarcoplasmic reticule. Many researches have been done to improve the tenderness of meat samples (Morgan et al., 1991; Wheeler et al., 1992; Steen et al., 1997; Pérez-Chabela et al. 1998; Aktas and Kaya, 2001; Berge et al., 2001), but results are limited to certain kind of muscles. On the other hand, marination could be employed to extend the shelf life of meat and increase the value of hard cuts with low commercial value, besides the activation of endogenous proteolytic enzymes, calpains. The most employed marinating method is immersion, with the concomitant impact of the time required (24-48 h) at low temperatures. Tumbling or massaging systems could be employed to reduce marination time since the massaging promotes a faster migration of solution into meat tissues.

Objectives

The objective of this work was to study the effect of calcium chloride marination of *brachiocephalicus* bovine muscle, a relatively tough muscle, in a tumbling system at different rpm, in order to reduce immersion time during meat marination.

Materials and methods

Bovine *brachiocephalicus* muscle (48 h post mortem) was obtained from a local abattoir. Meat was cleaned from visible connective tissue and fat, and cut perpendicularly to muscle fibers and randomly distributed in the different treatments, displayed in Table 1. In order to reduce marination times reported (Koohmaraie et al., 1990; Wheeler et al., 1997; Pérez-Chabela et al., 1998), four different tumbling levels were employed (i.e., 1000, 2000, 3000 or 4000 tumbling/min, obtained in 2, 4, 6 or 8 h, respectively), together with an immersion sample (48 h at 4-6°C), in a 150 mM $CaCl_2$ solution. An untreated sample was used as a control. Vacuum massaging was made employing a SVM-30C machine tumbling (Edel Ingenieros, Monterrey, Mexico), at –25 Hg in. Samples were vacuum packed and analysed 24 h after their respective treatments. Meat texture was determined in a TA-HDi texture analyser (Texture Technologies, Scarsdale, New York/ Stable Micro System, Surrey, England) equipped with a 50 kg load cell and a Warner-Bratzler device. Meat samples were compressed at 2 mm/s constant speed rate and the maximum force detected was reported as Warner-Bratzler shear force. Calcium chloride was determined colorimetrically with the AOAC Official Method No. 983.19 (AOAC, 1999). A total of three replications were processed and analysed. Results obtained were statistically analysed with ANOVA in the SAS Statistical Software v. 8.0 (SAS Institute, Cary, North Carolina). Significant differences between means was determined with Duncan mean test.

Results and discussion

Samples for T1 showed higher calcium chloride concentrations ($P<0.01$) than others (Table 2), probably due to the longer contact with $CaCl_2$ solution (48 h). Final calcium concentration was significantly different ($P<0.01$) between the treatments. Calcium chloride concentration was higher for immersion samples (0.91 mg Ca^{+2}/100 g), as compared to control (0.11 mg Ca^{+2}/100 g) or massaging samples (~0.70 mg Ca^{+2}/100 g in average). Control sample (T0) had the lowest calcium concentration. In the tumbling samples, no significant differences ($P>0.05$) were detected. It means that the calcium chloride absorption is independent of the number of tumbling per minutes, and marination time could be reduced until to 2 h instead the 48 h required in the immersion process. There was a significant difference in the meat tenderness, reported as WB shear forces values, by the treatment employed (Table 2). Calcium chloride had effect on meat tenderness (Morgan et al., 1991; Steen et al., 1997; Wheeler et al., 1997; Pérez-Chabela et al. 1998), activating proteolytic enzymes, calpains, which induce myofibrillar structure breakdown, since control sample was tougher than the marinated samples. In same way, no significant differences were detected ($P>0.05$) among marinated samples, despite the process employed.

Conclusions

Meat tenderness induced by calcium marination can be achieved with short massaging times (2-4 hours), where the calcium concentration achieved by tumbling was enough to promote calpains activation, modifying myofibrillar structure and improving meat tenderness in this relatively tough and low value muscle.

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Table 1. Experimental design of the treatments

Sample	Treatment
T0	Control
T1	Immersion 48 h at 4°C
T2	Tumbling, 1000 rpm
T3	Tumbling, 2000 rpm
T4	Tumbling, 3000 rpm
T5	Tumbling, 4000 rpm

Table 2. Duncan's mean test for shear force and calcium concentration results

Response	T0	T1	T2	T3	T4	T5
$CaCl_2$ concentration (mg Ca/ 100g)	0.12^{d}	0.91^{a}	0.64^{b}	0.74^{b}	0.70^{bc}	0.74^{b}
Warner-Bratzler shear force (kg)	26.45^{a}	14.20^{b}	14.01^{b}	13.67^{b}	11.57^{b}	11.48^{b}

[a, b] Means with same letter in same row are not significantly different (P>0.05)

DISTRIBUTION OF RELATIVE HUMIDITY IN DEPENDENCE OF THE INLET AIRFLOW DIRECTION IN DRY SAUSAGE RIPENING CHAMBERS

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Background

Due to quality and safety issues the ripening phase is the most critical stage in the production process of dry sausage. In contrast to the traditional ways of manufacturing, dry sausages nowadays are ripened and dried in modern multifunctional chambers under defined conditions (temperature, relative humidity, airflow velocity etc.) and a defined period of time, which depends on the product [1]. However, one of the main issues in the design and operation of modern ripening chambers is to achieve a homogeneous drying flow over the chamber length and height and knowledge about it's influence on the ripening process from an engineering point of view. The influence of different airflow directions on the local velocities at the sausages surface already has been discussed [2]. However, the mass transfer also depends on the water vapor partial pressure difference between the sausage surface and the surrounding air and is limited by the effective diffusion from the core to the surface [3]. The difference between the surface activity of water and the relative humidity (RH) of the surrounding air is the driving force of the drying process. To avoid drying errors this difference should not exceed 5 % during the whole process [4, 5, 6].

Objectives

In this project a method for quality maintenance in dry sausage production through the analysis of airflow and water transport in ripening chambers is investigated. Therefore, a method of measuring the relative humidity (RH) during the regular production in ripening chambers was developed. This study discusses the effect of different airflow directions on the distribution of the RH in a newly developed type of ripening chamber. Furthermore, the effect of natural convection on RH was investigated by cyclically interrupting the inlet air flow.

Materials and methods

Ripening chambers

The investigations were conducted in two different types (type 1 and type 2) of ripening chambers. Both chambers contained 60 trolleys (three in a row) loaded with 12 tons of the same sausage in total.

Conventional ripening chambers (type 1) work on a basic principle: Inlet air flow enters the chamber via two separate nozzles batteries, located at both sides of the ceiling. A rotary valve periodically varies the amount and ratio of air exiting the two feeding ducts. The inlet air flows descend vertically along the side walls and are diverted at the chamber bottom into a horizontal flow. Both jets merge over the bottom. The merging point is shifted periodically over chamber width by the variation of the ratio of the air inlet flows between both sides. In the following upward zone, the air stream flows through the sausages towards an exhaust channel located on the ceiling midsection.

In this flow type, dry air gets transported to the lower chamber areas with high velocity. Passing the sausages, the flow decelerates and due to water transfer from the sausages the saturation level of the humidity rises simultaneously. This results in an inhomogeneous drying over the height of the trolleys.

For this reason, a new type of ripening chamber (type 2) was developed (Ness & Co. GmbH, Remshalden, Germany), which uses two additional inlet nozzles batteries installed horizontally (Fig. 1). The airflow here is also distributed periodically between the two feeding ducts by a valve. The jets merge in the headspace and pass the sausages from the top to the bottom of the rack. Afterwards the air stream flows along the floor towards an exhaust duct located at the bottom of the left side of the chamber. Through a periodic change between these two flow types (vertical and horizontal), drying errors can be avoided.

Measurement of relative humidity RH

The RH and the temperature were measured with capacitive RH sensors (Model FH A646, Ahlborn, Holzkirchen, Germany). Eight sensors were fixed on a square steel bar at intervals of 20 cm (Fig. 2). This bar was attached in the middle of a trolley. The data were recorded by a data acquisition unit (Almemo 8990-8, Ahlborn, Holzkirchen, Germany). The trolley with the sensors was placed in different positions inside the chamber, in the case shown here, in the middle of the first row.

Results and discussion

Figure 4 shows a typical trend for RH values over the height of a trolley in chambers with vertical inlet flow. Sensor 1 was below the sausages, sensors 2-7 were arranged over the height, sensor 8 was in the head space. There has been a stratification from the bottom, with prevailing dry air from the climatisation, to the top, where the air has accumulated water from the sausages surface. The cyclical variations in the diagrams are due to the rotary valve in the feeding duct. Corresponding to the valves' position the RH values differ from 6 up to 12 %. This large difference causes an inhomogeneous drying within the trolley (Fig. 3). The sausages on the top of the trolley showed a bright reddish color, which indicates a low drying rate, in the lower positions the sausages were glaring red, caused by a high drying loss.

For the investigation of the relative humidity without forced convection, the fan has been stopped for 15 minutes. In this period the RH values increased asymptotically. RH should not be increased too much, otherwise a micro-climate will develop, where moulds and yeasts can grow on the sausage surfaces. The RH difference between the lower and the upper sensors has been reduced, because the values of the lower positions increased stronger (Fig. 5). After restarting the fan the RH values rearranged themselves quickly. Stopping the fan or reducing the fan speed cyclically can therefore be used in the later ripening phase to achieve a more homogeneous drying of the sausages and to save electrical energy. Figure 6 shows the influence of changing the inlet airflow direction from horizontal to vertical inlet airflow on the distribution of the relative humidity. After changing the inlet airflow direction at a run time of 25 min, the RH values of the lower positions increased while the values of the sensors on the top decreased. The result was a "cross-over" of the charted humidity values. So the change from horizontal to vertical inlet airflow direction had a positive influence of reducing the RH values differences over the trolley height. For this reason the periodic change from horizontal to vertical inlet airflow is an adequate means to achieve a homogeneous drying rate over the height in the whole chamber.

Conclusions

The inlet air flow direction in ripening chambers influences the drying rate, not only by convection but also by the distribution of relative humidity. In chambers with vertical inlet flow (type 1), the RH is stratified with the effect of an inhomogeneous drying over the trolley height. Cyclically interrupting the inlet air flow can be used to achieve a slightly better drying result. In newly developed ripening chambers (type 2) the problem of an inhomogeneous drying by RH layers has demonstrable been improved by a periodical change of the flow direction.

Acknowledgements

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Fig. 1: Newly designed ripening chamber with vertical and horizontal inlet airflow

Fig. 2: Arrangement of the RH sensors

Fig. 3: Trolley with inhomogeneous dried sausages

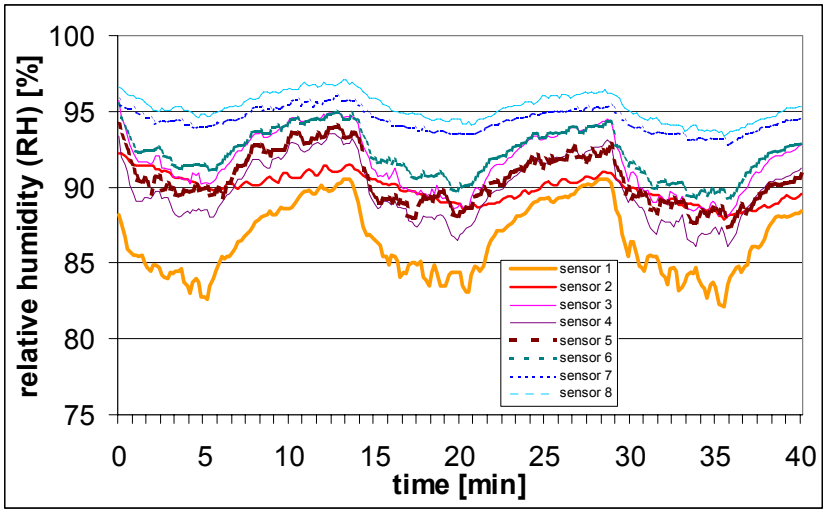

Fig. 4: Distribution of RH in chamber type 1

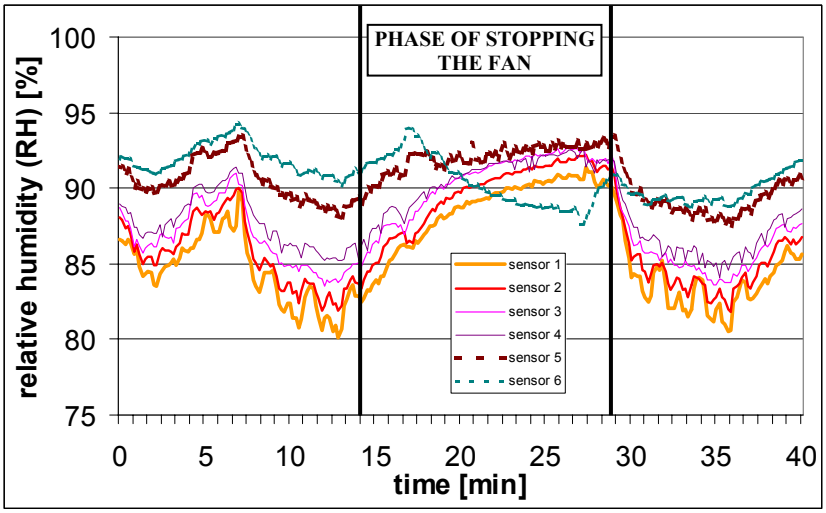

Fig. 5: Distribution of RH in chamber type 1 with stopped fan

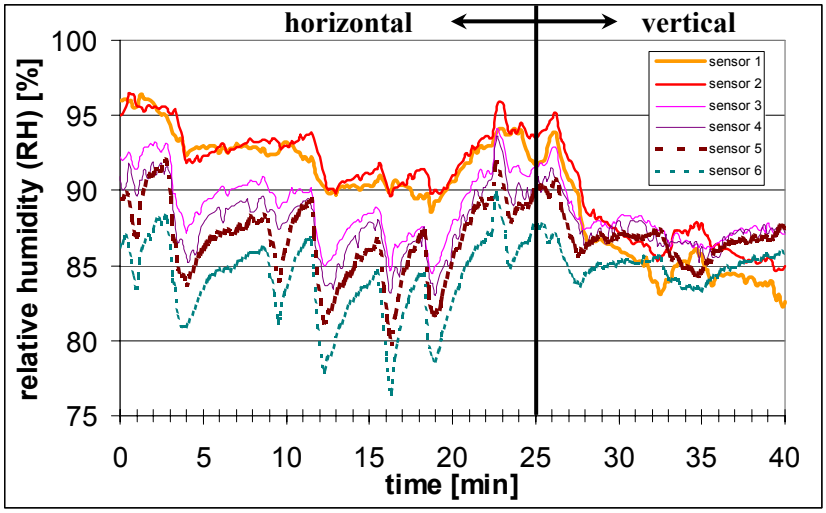

Fig. 6: Distribution of RH in chamber type 2 with change in the airflow direction

EFFECTS OF COOKING TEMPERATURE AND TIME ON GEL FORMATION ABILITY AND COLOR OF SURIMI-LIKE PORK

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Background and objective

Recently there has been considerable interest in manufacturing surimi-like materials (SLM) from the muscle of animal species. The characteristics of SLM from poultry meat, beef, pork, and sheep meat and also from meat by-products, such as beef or pig hearts have been studied (Kang et al., 2003; Lesiów and Xiong, 2003; Park et al., 1996). When the red meat is used to produce SLM, the high fat content of red meat, the more heme pigment and the high concentration of collagen cause several problems. Moreover, gel-forming properties are the most important attribute because surimi is formed and then cooked to make products. Several studies have been made to assess the effect of different temperatures on the gelation properties of SLM. Park et al. (1996) insisted that heating rate, endpoint temperature, protein level and source are important factors in developing gel texture of SLM. However, there is a little information on the gel forming characteristics of surimi-like pork. Understanding the gel forming mechanism of SLM by heating could increase the value of unpopular cuts or by-products of pork, increasing profitability. The objective of this study was to evaluate the effect of various cooking temperatures and times on gel properties of SLM made from pork.

Materials and methods

The *semimembranosus* pork muscle was obtained by hot boning, commercial vinyl pack-packed and stored during 3 days at cold room (2□4℃). After aging, removal of caps, vessels and external fat tissue, the lean muscle was diced into approximately 2 ㎝ cubes, and ground through a 4.7 ㎝ diameter orifice with a Kitchen Aid mince, then frozen at -60℃ until processed. The diced muscle was thawed at 4℃ for 24 hr. The lean muscle was chopped in a silent cutter with five volumes of iced water and the resulting slurry was filtered through a metal sieve of 1 mm mesh to remove connective tissue and filtered through in a metal sieve of 500 one more time. After filtered slurry was centrifuged for 15 min at 2,220 g at 4℃ and the supernatant discarded. The residue was re-washed with five volumes of water. A final wash was done in 2.5 volumes (v/w based on original weight of mince) of cold water. The washing procedure was repeated a third time. The resulting residue was centrifuged for 10 min 2,220 g at 4℃ and the supernatant discarded. For washed pork muscle, wet samples were stand upside down centrifuge bottle (500 ml) at cold temperature room for 10 min then removed free water of excess. Finally, SLM were re-mixed with 3% NaCl w/w, 0.5% tripolyphosphate w/w and 4% sorbitol w/w in a silent cutter for 4 min. The mixed SLM was stored in deep freezer at -60℃ and functional property by various cooking temperature were evaluated after 1 week. Stored SLM were kept overnight at 4℃ then stuff into a 62 mm diameter PVDC (polyvinylidene chloride) casing. SLM was heated for 20 min in a water bath at a constant of 65, 70, 75, 80 and 85℃, and heated at 75℃ for 15, 20, 25 and 30 min. Color (CIE L* and Hue) of cooked SLM was measured using a Minolta Chromameter CR-301 (Minolta Co., Japan) and Gel strength was measured with Sun Rheo Meter (COMPAC-100, Japan).

Results and discussion

The values of lightness, yellowness, chroma and hue were significantly ($p<0.05$) increased, whereas redness was significantly ($p<0.05$) decreased of cooked surimi-like pork by increasing of cooking temperature. pH and moisture % were significantly ($p<0.05$) increased as increasing of cooking temperature over 75℃. Also, hardness, springiness and gel strength were increased linearly as increasing of cooking temperature. Gel properties were evaluated by cooking times at cooking temperature 75℃. There was no significant difference in moisture % of cooked surimi-like pork among cooking time treatments. However, water-soluble protein solubility was decreased as increasing of cooking times. The values of lightness, hue and chroma were significantly increased with increasing cooking times, but lightness value was not changed over cooking of

25 min. Hardness and gel strength values of cooked SLM at cooking time of 15 min were significantly higher compared to cooking times over 20 min. However, panel evaluated the surimi cooked for 15 min as having a poor gel formation ability because it was too sticky.

Sarcoplasmic protein fraction pattern in SDS-PAGE showed that various enzymes were decreased as increasing of cooking times. The A band (phosphorylase) was remained at cooking of 15 min, but it was disappeared over 20 min cooking. The B (about 60 kDa) and D band (about 32 kDa) were remained until 30 min cooking, and they were dim at 35 min. Intensity of C band (about 46 kDa) was decreased at 20 min, and then it was disappeared. These results suggested that enzymes remained in sarcoplasmic could attribute to dark color and undesirable gel formation ability of cooked surimi-like pork. Especially it was assumed that approximately 46 kDa protein might be related to gel formation ability and color of cooked surimi-like pork.

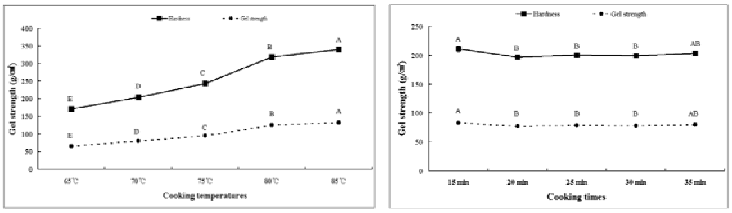

Fig. 1. Effect of cooking temperature and time on gel strength of cooked surimi-like pork.
Mean±S.E. $^{A-E}$Different letters are within a column indicate significant differences between mean values ($p<0.05$).

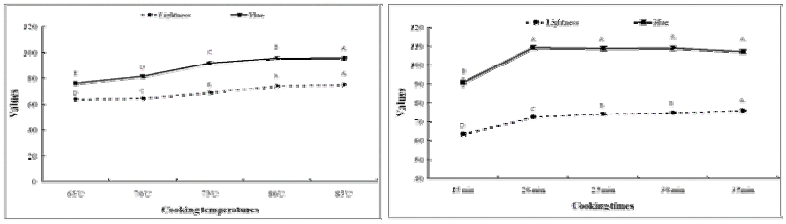

Fig. 2. Effect of cooking temperature and time on color values of cooked surimi-like pork.
Mean±S.E. $^{A-E}$Different letters are within a column indicates significant differences between mean values ($p<0.05$).

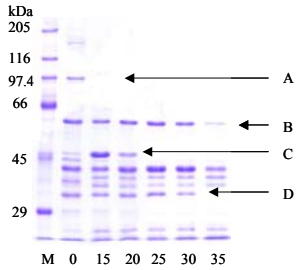

Fig. 3. Changes in protein fraction patterns of surimi-like pork by cooking times at at 75℃.

Conclusions

Gel strength and color of cooked SLM were improved as increasing of cooking temperature. Cooking time of 15 min at 75℃ was not enough for a desirable gel formation and color of SLM. Sarcoplasmic proteins that were not disappeared by cooking might be a reason for undesirable gel formation ability and color of cooked surimi-like pork.

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EFFECTS OF HIGH-PRESSURE TREATMENT ON INTRAMUSCULAR COLLAGEN MOLECULE

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Background

High-pressure treatment is an attractive application for foods, because that make possible non-heat food processing. High-pressure treatment is also one of the new technologies for tenderizing meat or accelerating meat conditioning (SUZUKI *et al*., 1998). Meat tenderness was determined by both actomyosin toughness attributed to myofibrillar proteins and background toughness attributed to connective tissue proteins mainly composed of collagen. It has been well known that high-pressure treatment causes the structural changes in a number of myofibrillar proteins. However, little is known about the effects of pressurization on connective tissue. We reported that intramuscular connective tissue was tenderized by high-pressure treatment and thermal stability of intramuscular collagen was decreased with pressure applied (ICHINOSEKI *et al.*, 2003). We also observed the structural weakening of perimysium and of perimysial-endomysial junction by high-pressure treatment.

Objectives

The purpose of this study was to confirm whether intramuscular collagen molecule could be degraded by high-pressure treatment. In this study, SDS-PAGE and differential scanning calorimetry of pepsin-soluble collagen and determination of collagen-derived peptide were investigated.

Materials and methods

Lean meat was removed from the shoulder of 76-86 months old Holstein cows 1 day after slaughter and stored at –20˚C. As required, it was tempered overnight in a cold room at 4˚C.

1. Isolation of intramuscular connective tissue

Intramuscular connective tissue was isolated by the method of FUJII & MUROTA (1982). After minced, muscle sample was homogenized in 10 mM Tris-maleate buffer, pH 7.2 containing 0.1 M KCl. Fibrous material in suspension was collected by passing the homogenate through a sieve with 1.0-mm pore. The residue was extracted with Hasselbach-Schneider solution and then 0.6 M KI-0.06 M $Na_2S_2O_3$. The insoluble residue containing collagen fibril was washed with distilled water. A part of the isolated connective tissue was lyophilized, and remainder was used for the following analysis.

2. Preparation of pepsin-soluble collagen

The isolated connective tissue was suspended in 0.5 M acetic acid containing 1 mg/ml pepsin and stirred overnight. The supernatant was collected by centrifugation at 100,000 *g* and precipitated by 2 M NaCl. After centrifugation, the pellet was resuspended in 50 mM acetic acid, extensively dialyzed against 5 mM acetic acid and then lyophilized.

3. Pressurization

Intramuscular connective tissue or pepsin-soluble collagen was packed in a polyethylene bag, sealed with cold distilled water and pressurized at 100-500 MPa for 5 min at about 8˚C using an isostatic press apparatus (Nikkiso KK, Tokyo).

4. SDS-PAGE of collagen

SDS-PAGE of collagen was carried out according to the method by HAYASHI & NAGAI (1979). Pepsin-soluble collagen was dissolved to a final concentration of 2 mg/ml in sample buffer (0.01 M Tris-HCl, 3.6 M Urea, 1% SDS, 1% β-mercaptoethanol, 0.01% BPB, pH 6.8). After heating at 100℃ for 3 min, the sample was subjected to SDS-PAGE analysis. Gels were stained with Coomassie brilliant blue R-250.

5. Differential scanning calorimetry (DSC)

Lyophilized pepsin-soluble collagen with or without high-pressure treatment was swollen in distilled water and then was put into the DSC stainless container. DSC was carried out using Setaram micro DSC VII at a heating rate of 0.5˚C/min and temperature range from 0 to 100˚C.

6. Determination of collagen-derived peptides

After pressurized, intramuscular connective tissue was suspended in twice volume of distilled water, and then centrifuged at 10,000 *g*. The supernatant was used for determining hydroxyproline content of collagen-derived peptides obtained by addition of trichloroacetic acid solution (final conc. 5%) (Fig.1). Hydroxyproline concentration was measured according to BERGMAN & LOXLEY (1963).

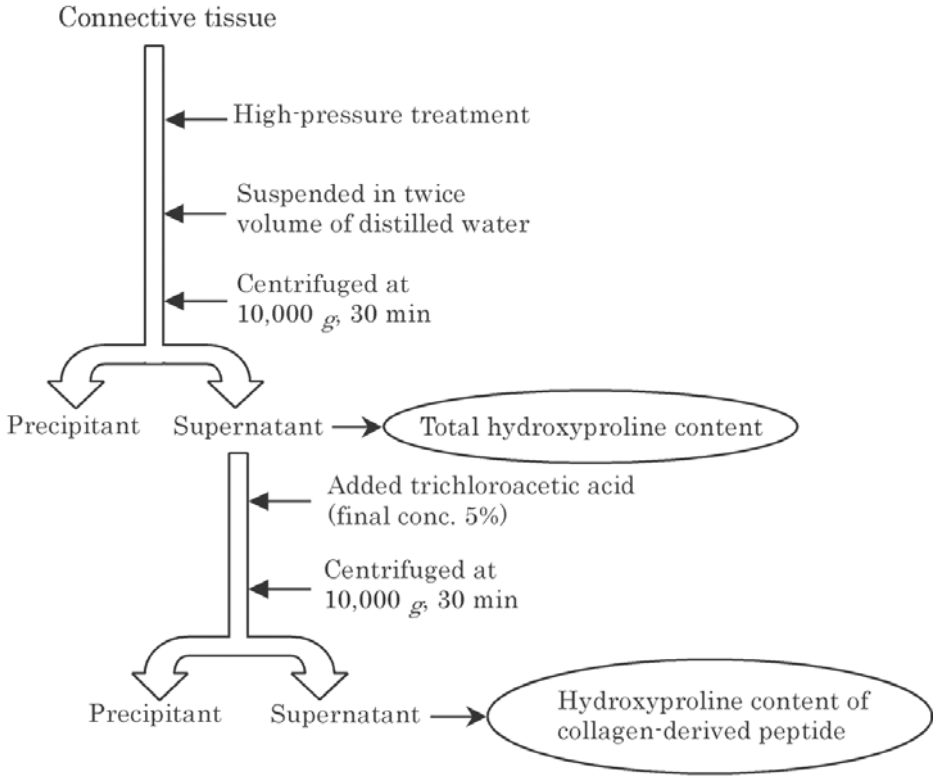
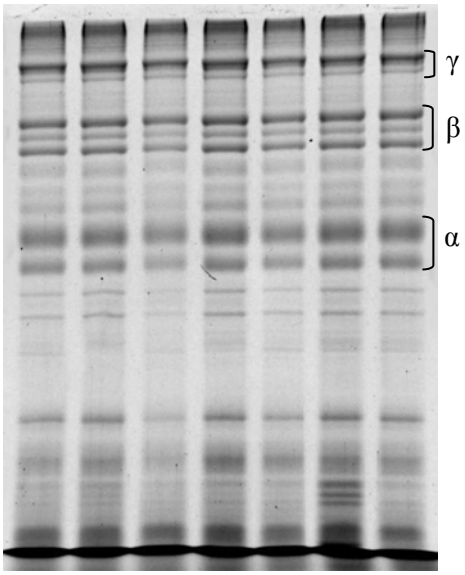


Fig.1 Scheme of determination of collagen-derived peptide.

Results and discussion

SDS-PAGE profile of pressurized pepsin-soluble collagen was shown in Fig.2. It observed that α, β and γ chains of collagen had no significant changes with pressure applied, which suggests that high-pressure treatment could not degrade collagen molecule.



(0.1) (100) (150) (200) (300) (400) (500)

Fig.2. SDS-PAGE profile of pepsin-soluble collagen.
(0.1) untreated, (100) pressurized at 100 MPa, (150) pressurized at 150 MPa, (200) pressurized at 200 MPa, (300) pressurized at 300 MPa, (400) pressurized at 400 MPa, (500) pressurized at 500 MPa.

Denaturation temperature of pepsin-soluble collagen was shown in Table.1. Pressure-induced significant changes in the denaturation temperature suggesting the denaturation of collagen molecule were not observed.

Table.1. Effect of pressurization on denaturation temperature of pepsin-soluble collagen.

Pressure applied (MPa)	0.1	100	300	500
Denaturation temperature (˚C)	42.9	42.7	42.0	42.3

Total hydroxyproline content in the supernatant (See Fig.1) increased with an increasing pressure applied, while hydroxyproline content of peptide fraction did not change. The proportion of collagen-derived peptide in exudation decreased from about 50% at 0.1 MPa (untreated) to about 20% at 400 MPa. These results suggest that high-pressure treatment could not degrade collagen molecular structure but could dissociate collagen super molecular structure, e.g. collagen fibers or fibrils, to collagen molecule.

Conclusions

From these results, it is suggested that the pressurization may cause some changes in the intramuscular collagen. The changes may not be involved in the degradation of collagen molecules but dissociation of collagen fibers or fibrils.

Acknowledgment

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TEXTURE PROPERTIES OF SAUSAGE EMULSIONS ANALYSED BY EXTRUSION METHODS AS PREDICTION INDEX OF SAUSAGE TEXTURE

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Background

In the last few years there has been a strong increase in the request and development of light products, that is, those of low-fat contents. In conventional cooked sausages, animal fats are an essential ingredient easily representing a 20-25% of the global composition. Reduction of the fat level has negative repercussions on the sensory quality of the final product, affecting mainly its texture.
The adequate combination of several compounds such as proteins, starches and hydrocolloids allow achieving a low-fat product with similar characteristics of texture the conventional product has. (Glicksman, 1991). There are a broad group of compounds with high functionality, but it is very important its adequate combination since sausage emulsion is a complex matrix where synergetic actions or complementation of properties of ingredients bring about (Dziezak, 1991; Keeton, 1994, Ordoñez et al., 2001).
In low fat product design, extrapolation of texture properties of final product from texture measurement of emulsion characteristics would be very useful. There are two instrumental methods to study semi-solid foods as emulsions, dough, jellies, etc. Both of them give a measurement of the compression force when product is extruded. Their differences are based on method of extrusion: Direct and back extrusion.

Objectives

The aim of this work was to find an easy instrumental measurement of sausage emulsion characteristics that can predict texture properties of final product, especially to be used in low fat sausages design.

Materials and methods

Emulsion texture analysis: Three different meat emulsions were used to establish analysis conditions. These emulsions differ with respect to protein and fat contents. Emulsion A was made with 13% protein and 13,3% fat, emulsion B 10% protein and 18% fat, emulsion C 8% protein and 13% fat. The rest of ingredients (water, additives, starch, etc) were added in the same amount to the three formulations.
The three experimental emulsions were evaluated by direct extrusion (probe A/BE) and back extrusion (probe HDP/FE5) with a texturometer Stable Micro Systems TA.XT2 (Haslemere, England). Maximum force along extrusion were taken and considered related to viscosity of emulsions. According to SMS Application Studies (1995), the maximum peak force (obtained from the F/t curve) is correlated with the viscosity of the product. Differences between both tests are based on how the product is extruded. Using the A/BE probe, the product is extruded up and around the edge of the disc plunger. However, with HDP/FE5 the product is extruded through a standard size outlet in the base of a sample container. Each data correspond to three measurements on 100 g emulsion at 20ºC in standardised containers. Compression percentages of 10, 15 and 20% and speeds of 0.8, 1 and 1.2 mms^{-1} were tested with both probes to determine the analysis conditions. Final conditions were 20% of compression and cross-head speed of 1 mms^{-1}. (Severiano Pérez, 2002).

Sausage texture analysis: Texture Profile Analysis (TPA) were performed with a probe of 50 mm of diameter using a Texturometer Stable Micro Systems TA.XT2 (Haslemere, England). Analysis conditions were level of compression of 55% with respect to initial height of sample and 1 mms^{-1} cross-head speed. Sausages, vacuum packed, were heated in water at 75ºC for 15 minutes. Samples were obtained by cutting off the ends of sausage, then the remaining sausage were divided into pieces of 10 mm length. Samples were held at 70ºC during instrumental evaluation. An average curve of consecutive measurements on ten portions of each sample were obtained. Texture parameters hardness, chewiness, adhesiveness, springiness and cohesiveness were deduced from the resulting force-time curve (Breene, 1975).

Sensory evaluation of texture was performed by a trained panel, constituted by twelve members. The selection and training of panel was done according to Severiano Pérez (2002). Texture attributes elasticity, hardness, chewiness, cohesiveness, adhesiveness, juiciness and fat perception were evaluated using a nine point scale. Samples were prepared identically than for instrumental analysis.

Experimental sausages: Five sausages were prepared with different fat contents and hydrocolloids added: carrageenan and xanthan gum and Genugel®, type CHP-200 (HERCULES, Copenhaguen, Denmark), a commercial mix of carrageenan and xanthan gum. Table 1 shows the content of these ingredients added with a view to modified emulsion properties and texture of light sausages. In all formulations, sausages were made with pork meat and fat and included 2% salt, 2% soya protein, 1.3% starch, polyphosphates, nitrite, ascorbic acid, flavourings and colouring.

Sausages were manufactured following a conventional process; sausage batter stuffed in 22 mm cellulose casings (Viscofan, Pamplona. Spain) was cooked until 72ºC internal temperature. Sausages were vacuum packed and kept under refrigeration until analysis in the same week they were manufactured. Two batches were made.

Results and discussion

First at all, it was verified there was no significant difference between batches for any parameter analysed. No differences were detected neither in both types of extrusion tests nor in sausage texture parameters.

The effect of reduction of fat content was remarkable on viscoelastic properties; emulsion A presented the highest force value compared with low fat emulsions (B, C, D and E). Therefore, decrease of percentage of fat lead to lower values of extrusion force, and this parameter could be considered an index of emulsion viscosity (Figure 1). The influence of fat level on emulsion viscosity could be determined both by direct extrusion and back extrusion. On the other hand, results of back extrusion show lower force of emulsion B (0.6% commercial mix Genugel®), than the other low fat emulsions. Emulsions C, D and E have very similar viscoelastic properties. Bigger differences were expected in emulsion characteristics according to previous studies (Severiano Pérez, 2002). However, there is enough variability to be able to study the possible correlation between textural properties of emulsion and texture of sausage.

Figure 2 shows mean values of instrumental texture parameters of the two batches. No significant differences between sausages were found in springiness, cohesiveness and adhesiveness. It was expected bigger differences between sausages, especially those made without hydrocolloids. Concerning to springiness and cohesiveness, these results do not agree with Mittal and Barbut (1993), who found higher springiness and cohesiveness in low-fat products (12-14% fat) than in conventional sausages (26% fat). Sausage A (conventional content of fat) is considerably harder and presents a higher chewiness than low fat sausages, being sausage B and E sausages with medium hardness and chewiness and sausages C and D are situated in the lowest extreme with respect to both parameters. There is disagreement in previous references about texture of low fat sausages and the effect of functional ingredients, with no modification of texture (Mittal et al., 1993, Bloukas and Paneras, 1996) or decrease or increase of hardness (Hand et al., 1989, Decker et al., 1986, Ordoñez et al., 2001).

Results of sensory evaluation of texture are similar to those found in instrumental measurement as it was expected. Parameters with significant variability between sausages were only hardness, chewiness and cohesiveness. Sensory texture profile follows the same pattern described in instrumental texture, corresponding the highest and lowest values of hardness and chewiness to sausages A and B, respectively, as described above. Sensory evaluation showed significant differences in cohesiveness, but the only two sausages clearly different are A and E. No differences between sausages were detected in elasticity, adhesiveness and juiciness.

Concerning to correlation between force resulting of extrusion tests on meat emulsion and sausage texture, irrespectively of method used (direct or back extrusion) significant correlations were found only with hardness and chewiness and cohesiveness sensory evaluated (Table 2).

Conclusions

Measurement of characteristics of emulsion by extrusion tests give an easy and quick estimation of texture of sausages after processing with respect to hardness and chewiness, two of the main constituents of texture. This measurement as an index of sausage texture is also useful in low fat products and results of both direct and back extrusion tests are comparable. However, further research on emulsions and sausages with bigger differences in other texture parameters as elasticity, adhesiveness and juiceness is necessary to determine the accuracy of extrusion force to predict a wide texture profile of sausages.

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Table 1. Sausage formulations. Percentages of ingredients differing in experimental sausages.

	Sausages				
Ingredients	A	B	C	D	E
Fat	22	14	14	14	14
Protein	12	12	12	12	12
Lactate	----	1.5	1.5	1.5	1.5
Genugel®		0.6	----	----	----
Carrageenan	----	----	0.4	0.2	----
Xanthan Gum	----	----	0.2	0.4	----

Table 2. Significant correlation coefficients between emulsion properties and texture parameters.

	Direct extrusion	Back extrusion
Sensory Hardness	0.690	0.685
Instrumental Hardness	0.542	0.596
Sensory Chewiness	0.736	0.766
Instrumental Chewiness	0.736	0.766
Sensory Cohesiveness	0.655	0.665

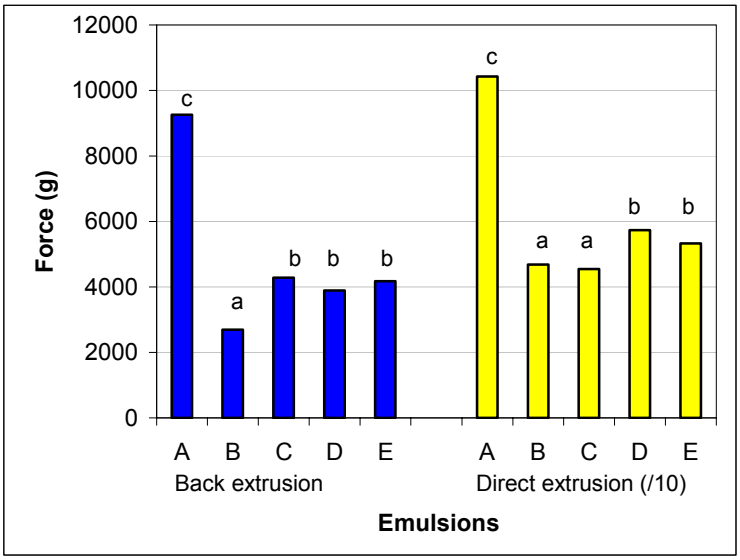

Figure 1. Evaluation of viscoelastic properties of different sausage emulsions (expressed as extrusion force). [a, b, c] For each type of extrusion columns with different letter are significantly different.

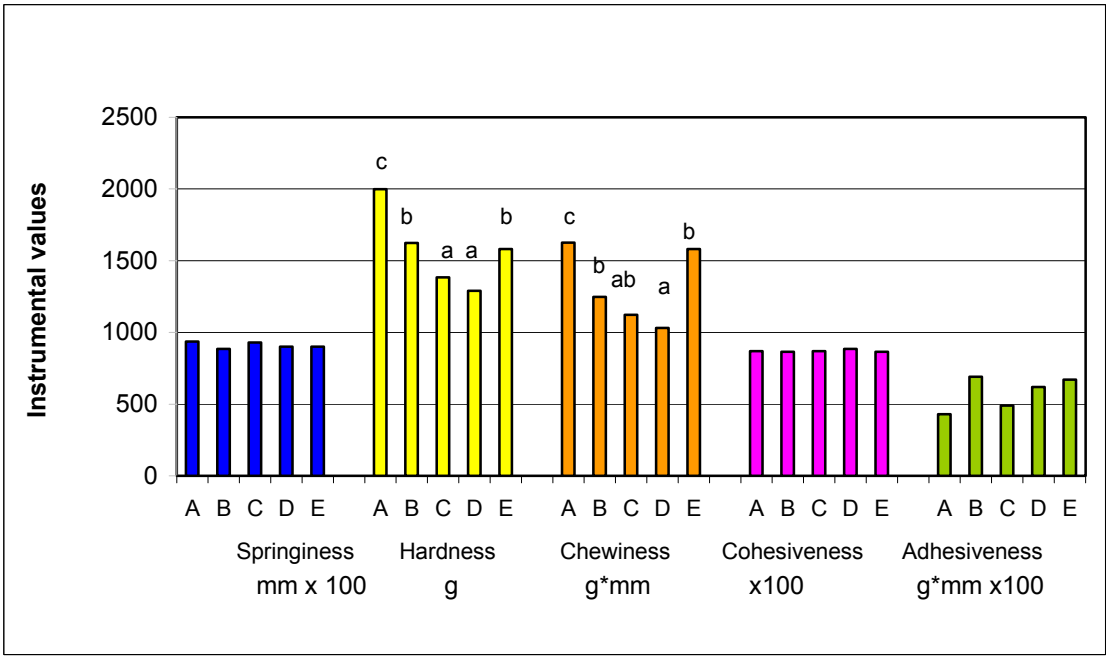

Figure 2. Instrumental evaluation of texture of experimental sausages. [a, b, c] For each texture parameter columns with different letter are significantly different.

GEL PROPERTY OF SURIMI-LIKE MATERIAL FROM PRE- OR POST-RIGOR PORCINE MUSCLE

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Background and objective

Surimi-like material from muscle of other animal species has been hypothesized to have similar properties to surimi from fish (Park et al., 1996). However, there is a little information on the preparation of surimi-like material from beef or pork. Pre-rigor beef is generally recognized to have superior functionally when used in the manufacture of processed meat products (Hamm, 1981). Salting of pre-rigor beef is known to promote maintenance of high binding quality (water and fat stabilization, texture development) during short-term refrigerated storage, presumably by solubilization the protein prior to a tight association of actin and myosin. Park et al. (1987) reported use of cryoprotectants to stabilize functional properties of pre-rigor salted beef during frozen storage. Also Park et al. (1993) reported cryostabilization of functional properties of pre-rigor and post-rigor beef by dextrose polymer and phosphates. The objective of this study was to evaluate the effect of rigor conditions of porcine muscle on gel properties of surimi-like material (SLM).

Materials and methods

Pigs were slaughtered at a Meat Plant of Gyeongasng National University, Jinju, Korea. The *semimembranosus* muscle was obtained by hot boning. After removal external fat tissue, the lean muscle was divided into three portions. For pre-rigor sample, one of the three portions was diced into approximately 2 ㎝ cubes, and ground through a 4.7 ㎝ diameter orifice with a Kitchen Aid mince, and SLM was manufactured. The others samples are commercial vinyl pack packaged and stored at cold room (2 ˚C 4 ˚C) until processed at *post mortem* 24 or 72 hrs. The *post mortem* 24 hr (rigor-mortis) and 72 hr (post-rigor) samples were also used for SLM manufacture, and the SLM manufacture procedure was modified to method of Park et al. (1996).

SLM yield % and color (CIE L*, Chroma and Hue) of SLM gel were measured using a Minolta Chromameter CR-301 (Minolta Co., Japan). Moisture content of uncooked and cooked SLM, and water-holding capacity (WHC) and water–binding capacity (WBC) were measured. Gel strength of cooked SLM was measured with Sun Rheo Meter (COMPAC-100, Japan) and SDS-PAGE was applied to investigate changes in sarcoplasmic and myofibrillar proteins of SLM.

Results and discussion

All SLM from muscles at pre-riogor, rigor-mortis and post-rigor was a light, opaque material with a dough like consistency. When additives (salt, TPP and sorbitol) were mixed with surimi-like pork, the material became comparatively clear and sticky. Specially, SLMs of pre-rigor and rigor-mortis muscles were lighter than that of post-rigor muscle after cooking. When pre-rigor muscle was used, more color was removed with the first washing step, resulting in lighter water washed material. After cooking of SLM, there were significant differences in Chroma and Hue values among treatments (Table 1). This color effect had been reported previously by Lan et al. (1993) and Park et al. (1996). However, yield % of pre-rigor muscle was sigificantly decreased compared to rigor-mortis and post-rigor muscles. Results suggested that sarcoplasmic proteins including pigments such as myoglobin and residual hemoglobin in pre-rigor muscle could be removed easily by water washing, resulted in having decreased yield of SLM. This was confirmed in sarcoplasmic proteins fraction of SDS-PAGE. Intensities of some sarcoplasmic enzymes such as phosphoylase were dim in rigor-mortis and post-rigor muscles (Fig. 2.). The sarcolasmic enzymes were still remained with myofibrillar proteins fraction in SLM.

Although WHC of SLM from post-rigor muscle was significantly lower (Table 2), gel strength and hardness of cooked SLM were significantly ($p<0.05$) stronger and harder in post-rigor muscle than those of other muscles (Fig. 2). Gel forming ability could be influenced by differences in WHC, WBC, protein concentration, ultimate pH and heating condition of SLM (Park et al., 1996). The lower moisture % in SLM

from post-rigor muscle might be related to gel strength and hardness. The lower moisture % in SLM implied that more concentration of proteins would be in SLM. Therefore, the harder gel of post-rigor muscle might be due to a higher concentration of protein in SLM without regard to the lower WHC. These results indicated that strong gel could be obtained with post-rigor porcine muscle because of higher concentration of protein in SLM. The post-rigor muscle, however, could produce dark color that would be avoid for surimi products. It was assumed that the dark color of SLM from post-rigor muscle might be due to a high concentration of sarcoplasmic proteins, oxidation of lipid and protein and the small space of gel matrix resulted in stronger absorption nature than reflection nature of light.

Table 1. Yield % and color of cooked SLM from different rigor conditions of porcine muscle

Muscle conditions	SLM yield %	CIE L*	Chroma	Hue
Pre-rigor	82.33 ± 7.42^{B}	78.51 ± 0.22^{B}	3.16 ± 0.07^{C}	159.61 ± 1.73^{A}
Rigor mortis	107.50 ± 2.17^{A}	80.30 ± 0.37^{A}	3.55 ± 0.07^{B}	126.96 ± 0.81^{B}
Post-rigor	100.77 ± 2.75^{A}	78.75 ± 0.21^{B}	6.61 ± 0.06^{A}	108.37 ± 0.70^{C}
SEM	3.79	0.18	0.17	2.34

Mean±S.E. $^{A-C}$Different letters within a column indicates significant differences between mean values ($p<0.05$).

Table 2. Moisture, free water, WHC and WBC % of SLM from different rigor conditions of pork

Muscle conditions	SLM moisture %		WHC %	WBC %
	Uncooked	Cooked		
Pre-rigor	89.10 ± 0.06^{A}	80.30 ± 0.70^{A}	64.61 ± 0.70^{A}	90.12±1.24
Rigor mortis	88.34 ± 0.93^{A}	81.33 ± 0.27^{A}	65.77 ± 2.32^{A}	91.53±1.05
Post-rigor	86.68 ± 0.31^{B}	77.18 ± 0.25^{B}	58.35 ± 0.83^{B}	88.51±0.70
SEM	0.36	0.43	1.01	0.63

Mean±S.E. $^{A-C}$Different letters within a column indicates significant differences between mean values ($p<0.05$).

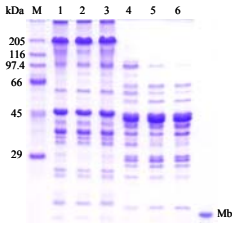

Fig. 1. SDS-PAGE patterns of obtained myofibrillar and sarcoplasmic proteins from rigor conditions of pork. (M : standard marker, lane 1: myofibrillar protein of pre rigor muscle, lane 2: myofibrillar protein of rigor-mortis muscle, lane 3: myofibrillar protein of post -rigor muscle, lane 4: sarcoplasmic protein of pre-rigor muscle, lane 5: sarcoplasmic protein of rigor-mortis, lane 6: sarcoplasmic protein of post-rigor muscle, Mb: horse heart myoglobin).

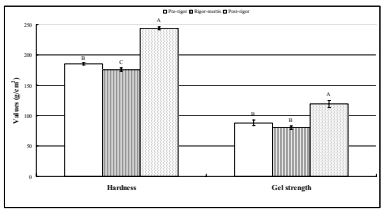

Fig. 2. Gel strength and hardness of cooked SLM from rigor conditions of pork. $^{A-C}$Different letters are within a column indicates significant differences between mean values ($p<0.05$).

Conclusions

A bright and white surimi could be obtained with pre-rigor porcine muscle, whereas a strong and hard surimi could be obtained with post-rigor muscle. However, post-rigor muscle can produce a dark color surimi and pre-rigor muscle can decrease yield % of surimi.

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METHOD AND DEGREE OF MECHANICAL TENDERISATION OF BEEF MUSCLES FOR USE IN RE-FORMED JOINTS MADE WITH A COLD-SET BINDER

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Background

The purpose of producing reformed beef products is to effectively market less valuable carcass components. These contain high levels of connective tissue and therefore require tenderisation for such products (Rolan et al, 1988). Tenderness is the single most important factor affecting consumers' perception of taste in beef (Morgan et al, 1991). Beef round and chuck are traditionally marketed in the form of low priced steaks or roasts. The present study is part of a project designed to increase the value of some muscles from these cuts through tenderising and re-forming treatments. Glitsch (2000) found that for European consumers, tenderness and flavour were the most important factors in eating quality of beef. Numerous reports have indicated that mechanical tenderisation reduces shear force values (Jeremiah et al, 1999). It has also been established that beef steaks and pork chops are more juicy and tender when injected with a phosphate and salt-containing solution (Robbins et al, 2003). Tumbling or massaging treatment is also used in the industry to increase tenderness. In this study several tenderising methods were assessed in conjunction with preparation of re-formed beef joints using Activa ™ (Ajinomoto Ltd.). The active ingredient in Activa is transglutaminase cross-bonding enzyme which catalyses the polymerisation and cross-linking of proteins (Kolle and Savell, 2003).

Objective

The objective was to compare the effects of three different methods of tenderisation on the processing characteristics and tenderness of two muscles selected from the beef chuck and two from the round.

Materials and methods

Beef muscles used were from the chuck (*M. pectoralis profundus* and *M. supraspinatus*) and round (*M. semimembranosus* and *M. vastus lateralis*) of R4L grade steers 4 days post-slaughter. *Semimembranosus* muscles were seamed out of the topside round and *Vastus lateralis* muscles were seamed out of the knuckle. The heavy central piece of connective tissue was removed from the *Supraspinatus* and the *Pectoralis profundus* was used whole. Muscles were trimmed of all visible external fat and connective tissue. Sufficient quantity of each muscle was pooled together to make twenty 19 x 13 x 7.5 cm joints (approx. 2kg weight) for steak cutting, a quarter being assigned to one of four treatments- 1) non-tenderised, 2) blade tenderised, 3) needle tenderised and 4) injected + vacuum-pulsed, giving five replicates of each treatment. For roasting joint production, which was an additional product made in the case of the injected + vacuum-pulsed treatment, five more such joints were made for each muscle group and these were compared with five commercial silverside round roasts. Blade tenderised (BT) muscles were passed once through a commercial roller-blade tenderiser. With this tenderiser, meat is passed between two rollers of 305mm length, with 78 blades per roller (blade width 1mm, 3.9mm interval between blades). Each blade contains 20 teeth of 9mm width with a 10mm interval space. Blades from opposing rollers overlap by 10mm. Needle tenderised (NT) muscles were passed once through a commercial Tender Star Model TSE tenderiser with solid stainless steel chisel-shape needles (needle diameter 1mm, at chisel end 2mm). The needle bank area of 250mm x 80mm contained 574 needles. Injected and vacuum pulsed muscles (VP) were injected with brine using a Dorit PSM-21 Inject-O-Mat brine injector at 16% to give a concentration of 0.5% salt and 0.3% sodium tripolyphosphate (STPP) in the meat, chopped to give pieces of ~200g weight and vacuum-pulsed for 14 hours in a Rühle vacuum tumbler/ mixer (Model MKR 150-600, Rühle GmbH) using a "Tender beef " programme which alternated between 90% and 10% vacuum (100 and 900 hPa). BT and NT muscles were also cut into ~200g pieces following treatment. Activa ™TG-RM at a level of 1% (% of raw meat weight) was whisked with 4% water until homogenous, and the suspension was then mixed with meat chunks, ensuring each piece was fully coated. Meat pieces were then layered in a polythene-lined rectangular-shaped mould (1.5-2.0kg per mould), vacuum packed and held at 2ºC overnight to allow completion of the protein

cross-linking reaction. Controls (CO) were treated in the same way but omitting the tenderisation step. The formed meat was removed from moulds and liners, and cut into steaks of 2.54cm width. Steaks were vacuum packaged and held at 2ºC pending analysis, with the exception of steaks for taste testing which were frozen at –20ºC for a minimum of one week. In the case of the injected and vacuum-pulsed treatment, joints for roasting were made in the same way but not cut up into steaks. They were then vacuum packaged in Cryovac cooking bags and following overnight holding cooked in a Jugema steam/ air cooker to an internal temperature of 70ºC using a cabinet temperature of 82ºC and a relative humidity of 99%. Five commercial round roasts made from the silverside, averaging 1kg in weight were cooked for comparison. Steaks were tested for cooking weight-loss, Warner-Bratzler shear force (WBS), colour (L*a*b*) and eating quality. Total viable bacteria counts (TVCs) at 30ºC were determined for steaks from one forequarter muscle (*Pectoralis*) and one hindquarter muscle (*Semimembranosus*) on day 0 and day 7. Cook loss was measured on the day steaks were cut and WBS on the following day. Cook loss was measured by weighing both prior to and after cooking to a core temperature of 70ºC. Shear values of 7 cores of 13mm diameter sheared perpendicular to meat fibre direction were measured. Colour was measured using a HunterLab spectrophotometer (Ultrascan XE) on the cut surface of PVC film-covered steaks after 1 hour blooming in air at 2ºC on the same day that steaks were cut. Measurements were taken on 10 locations per steak and averaged. Steaks for taste panels were thawed using cold running water. Taste testing was carried out on steaks grilled to a core temperature of 70ºC by a panel of eight people experienced in tasting beef. The panellists graded the samples for tenderness, chewiness, residual connective tissue, juiciness, overall flavour and overall acceptability on a scale of 1 (worst) to 6 (best).
Roast joints were tested for cook loss, texture (Kramer Shear Force), sliceability and eating quality. Cook loss was measured by weighing beef joints before and after cooking. Kramer Shear Force (KSF) was measured by placing approximately 30g of 1mm-thick roast beef slices in the bottom of a 10-blade Kramer Shear cell attached to an Instron model 4464 texture meter. Because of unavoidable weight variability between samples, measurements are expressed as Newtons force per g of meat sample. Sliceability was determined as the percentage of broken slices upon cutting 10 x 1mm-thick slices from a roast joint. Panels assessed taste and appearance of slices of the roast beef for colour acceptability, tenderness, juiciness, overall flavour, binding/ cohesion, overall acceptability and saltiness, on a scale of 1 (worst) to 6 (best). In the case of saltiness 1 represented not salty, 6 being extremely salty.
Analysis of variance of the results was carried out using the Genstat 5 Release 3.2 (Rothamsted Experimental Station). For total viable counts the analysis used was a split plot design.

Results and discussion

WBS results and taste panel ratings showed that type of tenderisation treatment significantly affected resulting steak tenderness (Table 1). For all except one muscle, CO samples, as expected, had higher WBS values than all other treatments, the exception being *Semimembranosus*, for which CO and BT samples were not different. Sensory panel tenderness ratings showed CO samples were significantly less tender than all treatment samples for 3 of the 4 muscles. Also in the case of 3 of the 4 muscles, BT had higher WBS than VP, and for both forequarter muscles, WBS values were higher for BT than NT. *Pectoralis* CO samples had higher WBS values than BT, NT and VP ($p<0.001$). This was also reflected in taste panel results where CO samples were rated less tender than BT and NT samples ($p<0.05$) and less tender than VP samples ($p<0.001$). CO samples were also rated more chewy than BT ($p<0.05$), NT ($p<0.01$) and VP ($p<0.001$) samples. Panellists also rated CO samples as having higher residual connective tissue content than BT ($p<0.05$), NT and VP samples ($p<0.01$). Overall acceptability ratings showed CO samples to be less preferable to NT and VP samples ($p<0.01$). BT samples had higher WBS values than VP samples ($p<0.001$), and were also rated less tender ($p<0.05$), more chewy ($p<0.01$) and less juicy ($p<0.001$) in sensory panels. *Supraspinatus* CO samples also had higher WBS values than all other treatments ($p<0.001$). This was reflected in sensory panel tenderness ratings for BT ($p<0.05$), NT and VP samples ($p<0.001$); chewiness ratings for NT and VP samples ($p<0.001$); residual connective tissue ratings for NT and VP samples ($p<0.01$) and overall acceptability ratings for NT and VP samples ($p<0.001$). BT *Supraspinatus* muscles had higher WBS values than NT and VP samples ($p<0.05$) and this was also reflected in tenderness, chewiness, residual connective tissue content and overall acceptability ratings for both muscles and in juiciness ratings for VP samples only. *Semimembranosus* CO samples had higher WBS values than NT ($p<0.01$) and VP ($p<0.001$) samples, but did not differ from BT samples. This corresponds to sensory panel ratings for tenderness, chewiness and overall acceptability, although panellists did differentiate between CO and BT samples, rating BT samples

better for tenderness, chewiness and overall acceptability. BT *Semimembranosus* samples had higher WBS values than VP samples ($p<0.01$) but this was not reflected in sensory results. *Vastus lateralis* CO samples had higher WBS values than BT, VP ($p<0.01$) and NT samples ($p<0.001$) but again, this was not reflected in sensory panel ratings. Overall, for all 4 muscles, VP samples were rated significantly more juicy than all others. They were also more tender in the case of all forequarter CO and BT samples, but panellists did not differentiate between these treatments in hindquarter muscles. WBS values also showed VP samples to be more tender than CO and BT samples for three of the four muscles. These results show that the superiority of the VP treatment over other tenderisation treatments used is evident in the forequarter muscles only, these having higher shear values than the hindquarter muscles in non-tenderised samples. These results correspond with a number of previous findings which indicated that brine-injection improves eating quality of beef e.g. Vote et al, 2000. Cook loss from steaks was not affected by treatments. Colour readings showed that for both forequarter muscles, NT samples were significantly lighter in colour, i.e. had higher L values than VP samples. There was, however, no difference in the lightness value between these two treatments in hindquarter muscles. *Supraspinatus* NT samples also had higher L values than BT ($p<0.01$) and CO samples ($p<0.05$). *Semimembranosus* CO samples also, had a lower a value than the corresponding NT samples (i.e. CO samples were less red than NT samples). Both hindquarter CO and BT samples had higher values than VP samples. Even though there were some differences in b values, no patterns emerged. From these results, overall, tenderisation treatments were not shown to have a negative impact on re-formed steak colour when compared to non-tenderised samples. Results of TVCs showed that for *Pectoralis* on day 0, the VP samples had a higher count than CO, BT and NT samples (VP=$5.1x10^5$ versus CO=$2.5x10^4$, BT=$7x10^4$, NT=$1.8x10^4$). On day 7, VP samples still had a higher count than all other treatments (VP=$1.7x10^6$ versus CO=$1.9X10^5$, BT=$4.9x10^4$, NT=$4.1x10^5$). Also, BT samples had higher counts than NT samples ($p<0.05$). *Semimembranosus* day 0 samples showed no differences between treatments, but on day 7 VP had higher counts than CO ($p<0.001$) and BT and NT samples ($p<0.01$). These results indicate that the injection and vacuum pulsing process allows for greater microbial contamination than the other tenderisation treatments used.

The comparison of roast beef slices from commercial silverside roast joints with those from the reformed joints showed (Table 2) that 3 out of 4 of the latter were as tender or more tender than the retail beef. Sliceability was better for reformed joints from all four muscles than for the commercial roasts. *Pectoralis*, *Semimembranosus* and *Vastus lateralis* reformed joints gave 100% sliceability and *Supraspinatus* gave 80%, while commercial silverside roast gave 67% sliceability. There was no significant difference in cook loss between commercial roasts and any of the reformed roasts.

Conclusions

All three tenderisation treatments used in this trial significantly reduced WBSF values, and in most cases these reductions were reflected in sensory panel ratings. Needle tenderisation and injection + vacuum-pulsing treatments gave about equal levels of tenderisation, while blade tenderisation had a lesser effect on tenderness. Injected + vacuum-pulsed samples retained more juiciness than those from other treatments for all four muscles. Tenderisation treatments were shown not to impair colour of re-formed steaks. Total viable bacteria counts indicated that injection + vacuum-pulsing results in higher counts than other treatments. Texture analysis of roast beef joints indicated that the commercial silverside roasts used for comparison, along with the *Pectoralis* roasts were both less tender than those from the other three muscles. The study indicates that needle tenderisation and injection + vacuum pulsing treatments are more effective than blade tenderisation for tenderisation of beef for use in reformed joints. Account needs to be taken of the possible higher bacterial numbers arising from the injection + vacuum-pulsing treatment.

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Table 1. Effect of tenderisation treatments on shear force values and taste panel ratings of re-formed steaks.

	Pectoralis profundus				*Supraspinatus*				*Semimembranosus*				*Vastus lateralis*			
	CO	**BT**	**NT**	**VP**	**CO**	**BT**	**NT**	**VP**	**CO**	**BT**	**NT**	**VP**	**CO**	**BT**	**NT**	**VP**
WBSF *	80.8^{a}	61.8^{b}	46.1^{c}	40.4^{c}	54.6^{a}	37.7^{b}	30.7^{c}	31.0^{c}	49.2^{ab}	44.3^{b}	38.4^{bc}	33.7^{c}	47.2^{a}	34.5^{b}	32.8^{b}	35.7^{b}
Tend.**	1.73^{a}	2.88^{b}	3.08^{b}	4.03^{c}	2.38^{a}	3.25^{b}	4.70^{c}	5.03^{c}	2.85^{a}	4.22^{b}	4.38^{b}	4.93^{b}	4.00	3.93	4.90	4.55
Chew.$^{\#}$	1.73^{a}	2.55^{bc}	2.93^{cd}	3.70^{d}	2.48^{a}	3.08^{a}	3.88^{bc}	3.93^{c}	2.78^{a}	3.70b	3.85^{b}	4.10^{b}	3.43^{a}	4.05^{ab}	4.08^{ab}	4.52^{b}
RCT §	2.28^{a}	2.95^{b}	3.28^{b}	3.48^{b}	2.83^{a}	3.08^{a}	4.08^{b}	4.00^{b}	3.45	4.22	4.18	4.07	4.18	4.43	4.70	4.13
Juicin. ¤	3.43^{a}	3.38^{a}	3.60^{a}	4.90^{b}	3.85^{a}	3.58^{a}	4.05^{a}	5.25^{b}	3.78^{a}	3.95^{a}	3.80^{a}	4.90^{b}	4.00^{a}	2.05^{b}	3.90^{a}	5.16^{c}
O. acc.«	2.35^{a}	3.03^{ab}	3.35^{b}	3.45^{b}	2.93^{a}	2.90^{a}	4.15^{b}	4.25^{b}	3.25^{a}	3.73^{bc}	4.18^{d}	4.00^{cd}	3.95^{ab}	3.50^{a}	4.18^{b}	4.31^{b}

a-d Treatment means with different superscripts, for each muscle, are significantly different ($p<0.05$)
* Warner Bratzler Shear Force (N)
** Taste panel tenderness
# Taste panel chewiness
§ Taste panel residual connective tissue
¤ Taste panel juiciness
« Taste panel overall acceptability

Table 2. Comparison of shear force values and taste panel results of vacuum-pulsed and injected beef muscles with silverside round roast

	Silverside round roast	**Pectoralis**	**Supraspinatus**	**Semimembranosus**	**Vastus lateralis**
KSF *	69.5^{a}	62.1^{a}	49.7^{b}	47.7^{b}	49.9^{b}
Tend. **	4.70	4.65	4.65	4.38	4.50
Juicin. $^{\#}$	4.33	4.38	4.48	4.53	4.65
O. acc. «	4.10	4.35	4.43	4.08	4.53

a-b Treatment means with different superscripts, for each muscle, are significantly different ($p<0.001$)
* Kramer Shear Force (N)
** Taste panel tenderness
# Taste panel juiciness
« Taste panel overall acceptability

HEAT-INDUCED CHANGES IN THE MECHANICAL PROPERTIES OF PERIMYSIAL CONNECTIVE TISSUE FROM TWO BEEF BREEDS

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Background

The structural origins of variations in meat tenderness, arising from various treatments or animal characteristics, are far from clarified. Results from macroscopic mechanical methods can lead to conflicting conclusions. Micro-mechanical tests performed on isolated structures (Lewis and Purslow 1989) allow clear understanding of the contribution of theses structures to the overall meat tenderness. Theses techniques have been used to elucidate the mechanical changes in connective tissue and muscle fibres associated with contraction state, ageing or cooking conditions (Christensen, Purslow and Larsen 2000). Only few data are available on the variations in the mechanical properties of these structures associated with different animal characteristics.

Objectives

The purpose of this study was to quantify heat induced changes, in mechanical properties of isolated perimysium sheets from beef muscles from two breeds.

Materials and methods

Muscles : *Semimembranosus* muscles were taken from cows (6 -7 years old) Holstein (12) and Salers (12). The muscles were removed from each carcass at 24h *post-mortem*. They were divided into 2 parts which were vacuum packed and stored at 4°C for 14 days and then frozen. One part was for measurements on raw meat and the other for measurements after 90 min cooking at 70°C.

Measurements :
All the samples were thawed in water at 10°C.
Measurements on raw meat:
The resistance of muscle fibres was measured in compression according to the method of Lepetit and Buffiere (1995) using an Instron 4501 testing machine. Sheets of perimysium (approximately 10 mm long x 4 mm wide) were dissected and tested with a micro-tensile device described below, for the determination of breaking stress, breaking strain, Young's modulus, breaking energy and total energy. The width of the samples were determined using a microscope and the thickness determined with a Mitutuyo micrometer under a force of 0.2N applied on the whole surface of the sheets. During the tensile test the samples were immersed in meat drip.
Measurements on cooked meat :
The cooking was done in water bath on 10x10x4 cm samples, the length in the direction of muscle fibres being 10 cm. Cooking losses were determined. The breaking stress of cooked samples was measured in compression with an Instron 4501 (Lepetit, Grajales, and Favier 2000). Sheets of perimysium (approximately 10 x 4 mm) were dissected from cooked meat and tested with the micro-tensile device. Width and thickness were measured as above. During the tensile test the samples were immersed in the liquid lost during cooking.
Micro-tensile test :
Tensile test was carried out on a micro-tensile device developed in this laboratory (figure 1). The procedure for handling samples is similar to that described by Lewis and Purslow (1989). After dissection, strips of perimysium were glued on aluminium foil frames with cyanoacrylate glue. The samples on the aluminium frames were then fixed on the micro-tensile device so that the direction of tensile testing was in the direction of collagen fibres.
The extension rate was 130 µm/s. The software applied a slack toe correction to the force – displacement curves to remove the part of the displacement where collagen fibres are just unfolded. The actual lengths of

the samples were determined from the rapid increase in force which happened when the perimysium structure entered into tension. The Young's modulus was calculated as the slope in the straight region of the stress – strain curves.

Statistical analysis :
For all mechanical variables means were obtained from 10 measurements. Data were analysed using the general linear model procedure of SAS Software (SAS/Stat Cary, NC: SAS Institute Inc., 2000).

Results and discussion

Meat from both breeds was not fully aged after 14 days of storage at 4°C as the resistance of muscle fibres were higher than the limit of 4 N/cm2 (Lepetit and Buffiere 1995) as seen in Table 1. Maximum compression stress of raw meat did not show any differences between breeds, but drip was significantly higher in meat from Salers than in Holstein. The breaking stress of cooked meat from Salers was significantly higher than from Holstein and cooking losses were similar between breeds (Table 2). The mechanical properties of perimysium sheets in the raw and cooked states are given in Table 3. The breaking stresses of perimysium sheets did not differ between breeds when compared raw or cooked, but there was a 36% reduction of the mean stress by cooking for 90 min at 70°C. The breaking strain was similar for both breeds in raw samples. For cooked samples breaking strain was slightly higher in Holstein than in Salers. Breaking strain increased, on average by 31% by cooking. Young's modulus decreased by 52% from raw to cooked. Breaking energy showed no significant variation neither between breeds nor from raw to cooked, whereas the total energy for the disruption of perimysium sheets showed a 25% reduction by cooking. The values of drip found for both meats (Table 1) include the drip occurring during 14 days of ageing and also the exudation due to thawing which explain why these values are quite high, but they are in agreement with values given by Offer and Knight (1988). The cooking loss agrees with values observed previously for the same muscle cooked in same conditions (Lepetit, Grajales and Favier 2000).

The breaking strain of perimysium sheets increased with cooking as collagen is progressively denatured and becomes rubber-like (McClain, Kuntz and Pearson 1969). Our values of breaking strain are much lower than those found by Lewis, Purslow and Rice (1991) for two reasons. First, in the present study, a slack toe correction was applied to the force-displacement curves to remove the displacement which corresponds to the unfolding of the collagen fibers. Secondly, the perimysium sheets in the present study, are cut parallel to one of the main directions of the collagen ply whilst samples in the study of Lewis, Purslow and Rice (1991) were cut perpendicularly to the direction of muscle fibers. Under those conditions, there is not only an unfolding of collagen fibers but also a reorientation of collagen fibers in the direction of the strain during the tensile test. In our samples no reorientation of collagen fibers occurred because they already are in the direction of strain and therefore a lower breaking strain is expected.

Values of the breaking stress from the present study are about four times higher than those reported by Christensen, Purslow and Larsen (2000) but here also precise comparisons are difficult due to the different type of samples. The present study concerned perimysium sheets from cull cows (6 – 7 years old) whereas the previous study concerned perimysium from young (2-2 ½) heifers and the resistance of connective tissue changes significantly with the age of animal as shown by adhesion measurements (Bouton and Harris 1972). Also differences in breaking stresses between the two studies may come from differences in sample shape and collagen fibre direction which both affect the number of collagen fibres contributing to the stress. Comparisons of Young's moduli lead to similar conclusions as for the breaking stress as they are highly correlated ($r=0.93$, $n=24$, $P<0.01$ for raw samples ; $r=0.87$, $n=24$, $P<0.01$ for cooked samples). Breaking energy, which represents the work done up to the maximum stress, was not affected by cooking. This results from opposing variations of breaking strain and breaking stress with cooking. There is a decrease in breaking stress and an increase in breaking strain due to cooking which lead to an almost constant breaking energy from raw to cooked. The total energy needed to separate perimysium sheets was significantly reduced by cooking. The energy which is developed after breakage is due to shear between collagen fibres. It represents about 60% of the total energy in raw samples and about 45% in cooked samples. It is this energy of shear which is decreased by cooking. In a fibrous composite material the energy of shear is supported by the matrix, which, in the case of perimysium, is composed of proteoglycans.

The maximum stress of compression of raw meat reflects mainly variations in connective tissue and therefore was not expected to vary between breeds as the data on perimysium sheets did vary between breeds. The small differences between breeds in maximum stress of cooked meat could not be linked to the minor variations between breeds in mechanical properties of cooked perimysium sheets.

Conclusions

Tensile tests on perimysium sheets isolated from *semimembranosus* muscles of cows from two breeds show significant changes with cooking (90 min at 70°C) in breaking stress, breaking strain, Young's modulus and total energy. On raw perimysium sheets no differences between breeds was observed which is in agreement with compression data on raw meat. The small differences between breeds in maximum compression stress of cooked meat cannot be linked to the minor variations in mechanical properties observed on cooked perimysium sheets.

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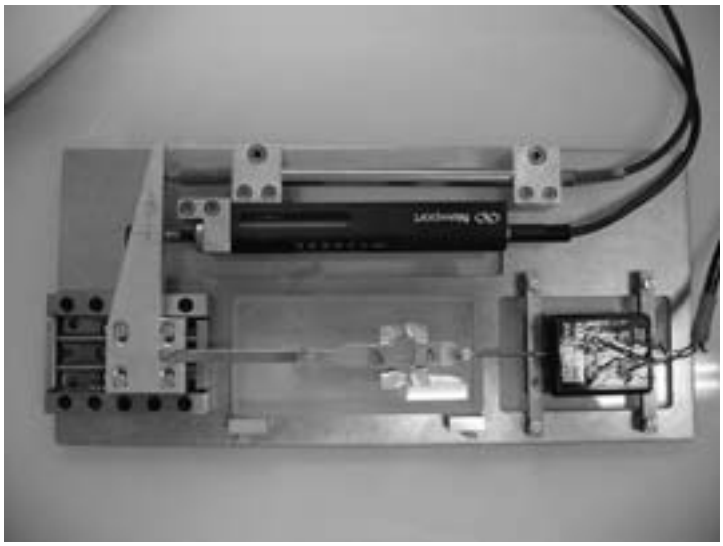

Figure 1 : Micro-tensile device. Total length 28 cm.

Breed	Resistance of muscle fibres (N/cm2)	Maximum stress (N/cm2)	Drip (%)
Holstein	8.1 *a*	85.1 *a*	7.6 *a*
Salers	7.0 *a*	82.5 *a*	8.6 *b*

Table 1 : Mechanical properties and drip of raw meat
In each column the values followed by different letters are significantly different at a 5% level.

Breed	Maximum stress (N/cm2)	Cooking loss (%)
Holstein	204.7 *a*	21.8 *a*
Salers	226.8 *b*	21.2 *a*

Table 2 : Mechanical properties and loss of cooked meat
In each column the values followed by different letters are significantly different at a 5% level.

	Breed	Breaking Stress (MPa)	Breaking strain	Modulus (MPa)	Beaking energy (mJ)	Total energy (mJ)
Raw	Holstein	10.3 *a*	0.36 *c*	41.0 *a*	8.5 *a*	20.0 *a*
Raw	Salers	12.5 *a*	0.35 *c*	52.4 *a*	8.6 *a*	20.5 *a*
Cooked	Holstein	7.6 *b*	0.49 *a*	22.1 *b*	9.2 *a*	15.9 *b*
Cooked	Salers	7.1 *b*	0.44 *b*	22.4 *b*	7.9 *a*	14.5 *b*

Table 3 : Mechanical properties of raw and cooked perimysium sheets
In each column the values followed by different letters are significantly different at a 5% level.

DEVELOPMENT OF TECHNOLOGY FOR PRODUCTION OF MEAT-PLANT EXTRUDATES

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Background

Extrusion technology is one of the advanced methods for obtaining high quality foods. The range of products manufactured using this technology includes more than 400 items: dry breakfasts, products for nutrition of children, cracker-type bread, potato chips, corn and wheat flakes, etc. /1/.
The advantages of extrusion technology consist in flexibility of its technological systems, continuity of the process, possibilities of simultaneous homogenization of raw materials and their thermal treatment which allow to solve many important problems in foods production, i.e. : preservation of biological and food value, inhibition of development of pathogenic microflora, formation of prescribed structural and mechanical properties and organoleptical characteristics.
These advantages indicate good prospects of use of extrusion technology in meat industry for development of meat-plant extrudates. It should be noted that the problems associated with peculiarities of formation of structure in the process of thermoplastic extrusion have been little studied.

Objectives

The purpose of this work was to investigate the influence of technological parameters of extrusion process (temperature, frequency of screws rotation) on structural and mechanical properties and quality characteristics of meat-plant extrudates.

Materials and methods

As objects of investigations were chosen products of animal origin: beef of 2nd grade, dry animal broth, light food grade albumin; of vegetable origin: corn starch and wheat flour. Second grade beef was comminuted (d = 2-3 mm) and mixed with other components in different ratio: 1) 2^{nd} grade beef, dry animal broth, corn starch; 2) 2^{nd} grade beef, wheat meal, corn starch; 3) 2^{nd} grade beef, light food grade albumin, corn starch. The mixtures were maintained during 12 hours at 4^0C for moisture level stabilization. Extrusion treatment was carried out on an experimental laboratory double screw extruder with the selected profile of configuration of screw elements of shafts for treatment of meat containing mixtures with the capacity of 40 kg/hr.
During investigations standard methods of determination of microstructure, mass fraction of moisture, fat, protein, carbohydrates were used. Digestibility of meat-plant extrudates was determined by the method (2). Structural-mechanical properties of extrudates were determined on the INSTRON - 1140 apparatus. Bulk mass p (kg/m^3) was determined by weighing in the capacity 1000cm^3. Coefficient of explosion $K_в$ was found as the ratio between bulk weights of mixture and final product.

Results and discussion

Quality of final products obtained as a result of extrusion treatment of selected recipe mixtures depends on quality of raw materials and parameters of the process: temperature of treatment of mixture and rotation frequency of screws.
Analysis of influence of extrusion treatment temperature shows that with temperatures <150 ^{0}C the extrusion process doesn't reach completion (Fig.1): biopolymeric mixtures don't melt to the end. At the exit from the extruder matrix there is no explosion of starch grains; the product is low-porous with low coefficient of explosion. Increase in the temperature from 150 to 180^{0}C results in $K_в$ increase; denaturation of native proteins and gelatinization of starches occur. In this case crystalline areas of biopolymers melt and the amorphous ones change from disordered high elastic state to a viscous-flow one, and as a result of sharp pressure drop - "decompression shock" - the extrudates have good porous structure /3/.
Temperature increase to higher than 180^{o}C leads to a decrease of the coefficient of explosion. This can be explained by more intensive Maillard reaction, the extrudates are strengthened, their porosity is reduced, and

hence, the coefficient of explosion is reduced, the extrudates acquire pronounced dark color and bitter off-flavor.

The investigations of the influence of rotation frequency of screws on extrudates quality have shown (Fig.2) that with low values of rotation frequency (12.7 s^{-1}) the shear stress of extrudates is not large. This occurs because the mixture is for a long time in the chamber of the extruder and undergoes strong destructive changes.

Proteins are destroyed to amino acids, polysaccharides to dextrins which interact forming different complexes. However, as a result of long effect of temperature, "block-dextrin" complexes are destroyed which results in low value of shear stress.

When the rotation frequency of screws increases to 13.7 –14.7 s^{-1} destruction of complexes "protein-dextrin" takes place not so intensively, but rather strongly. As a result the product is stronger, and the shear stress increases.

If the extrusion process is carried out with frequency rotation of screws 15.7 s^{-1}, the obtained extrudates are porous, easily crumble, don't have burned flavor.

With frequency rotation higher than 16.7 - 18.7 s^{-1}, the extrusion mixture is subjected to the effects of large shear characteristics with the result of destruction of mixture components, and hence shear stress increases.

The obtained extrudates (Table 1) are the products with high level of protein (8.9 – 15.9%) and low level of fat (0.35 – 0.58%).

The investigations on digestibility of these products have shown high degree of availability of proteins for proteases, which indirectly proves a deep destruction of proteins of meat-plant extrudates as a result of thermoplastic extrusion.

Investigations of microstructure of the comminuted extruded product (Figs. 3,4) (x 12) made it possible to establish that the main mass of particles in extrudates has plate-like corrugated structure and flowing internal structure. Methylene dye has clearly determined linear arrangement of protein component in the volume of the investigated particles. However, during the investigation mutually perpendicular orientation of protein and carbohydrate components was revealed. Muscle fibers that occur in the product consist of destroyed bundles of myofibrils with lost sarcolemma; there was a cross striation in some of their parts.

Conclusions

Thus, the experiments have shown that the technology of thermoplastic extrusion is suitable for manufacture of new kinds of meat products. To obtain extrudates of required quality meat-plant mixtures should be treated at technological regimes, as follows: matrix temperature – 180^0C, rotation frequency of screws – 15.7 s^{-1}.

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Table 1. Chemical composition of meat-plant extrudates

Index, %	Extrudate		
	1	2	3
Moisture	8.9	7.8	7.7
Protein	8.9	8.6	15.9
Fat	0.35	0.58	0.40
Carbohydrates	77.65	81.22	74.4
Ash	4.2	1.8	1.5
Digestibility, %	77.1	74.89	78.76

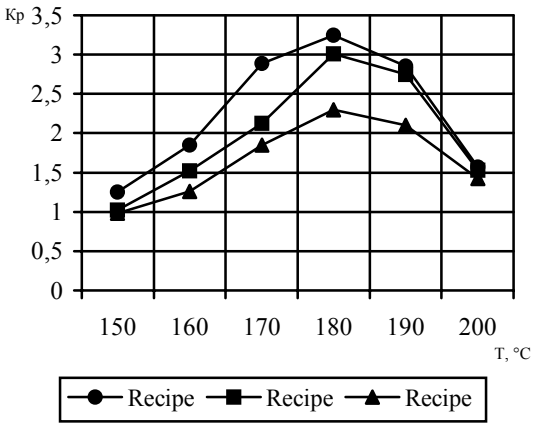

Fig.1 Dependence Кв from temperature of treatment at screw rotation frequency 15,7 s^{-1}

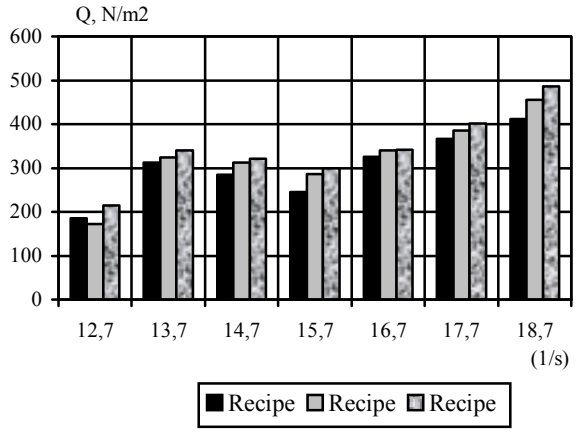

Fig.2 Dependence of shear stress Q ($N/м^2$) from screw rotation frequency at 180 °C

Figure 3. Microstructure of the comminuted extruded product. (x12)

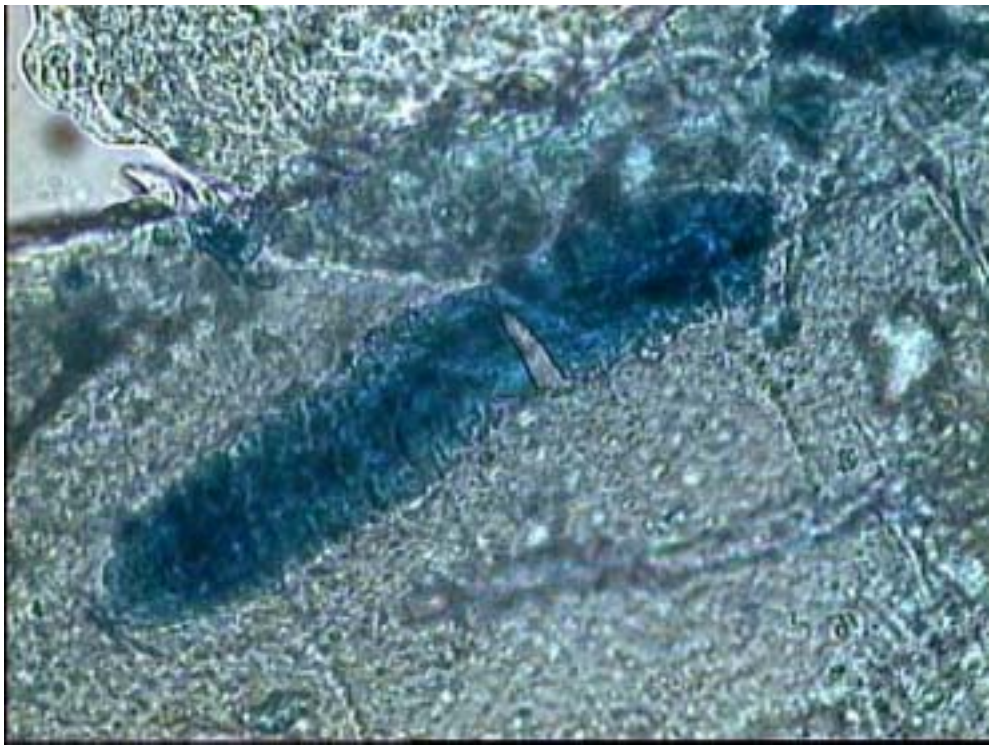

Figure 4. Microstructure of the comminuted extruded product. (x12)

INFLUENCE OF ANATOMICAL ORIGIN OF RAW MEAT ON THE SENSORY AND CHEMICAL CHARACTERISTIC OF DRIED BEEF "CECINA DE LEON"

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Background

Spanish "Cecina" is a salted, dried, and smoked beef meat product manufactured traditionally in the province of Leon (north-western Spain). It is an intermediate moisture product meat, made from three possible different anatomical retail cuts: "babilla" (composed of by *M. rectus femoris* and *vastus lateralis*, *vastus. medialis* and *vastus intermedius*), "tapa" (M. s*emimembranosus*, *sartorius*, g*racilis*, *adductor*, *pectineus*, *quadratus femoralis* and the external extra pelvic portion of M. *obturatorius externus*) and "contra" (*M. semitendinosus* and g*luteobiceps*). The preparation method is similar to that used in dry-cured ham manufacture. The final product has a typical red colour, smoked flavour and a characteristic slight salty taste. "Cecina" is highly prized in Spain and has recently been exported to the rest of Europe. Despite this, few studies have been carried out on the "Cecina de Leon".

Objectives

The objective of this research was to evaluate physicochemical and sensory characteristics of the three different types of "Cecina de Leon" ("babilla", "tapa" and "contra").

Materials and methods

Ten pieces of "babilla", ten of "tapa" and ten of "contra", provided by Protected Geographical Indication (PGI) "Cecina de Leon" were analyzed. All samples were vacuum packed and refrigerated at -4ºC until analyses. The analytical determinations were carried out in triplicate and the results expressed as dry matter.

The samples were subjected to the following analyses: pH, water activity (a_w), moisture, protein, fat and ash content. These physicochemical parameters were determined following the Official Methods of Analysis (BOE 29/8/1979) and the ISO recommended methods (ISO R-1442, ISO R-937, ISO R-1443, and ISO R-936 respectively). The following analyses were also performed: total carbohydrates (Luff-Schoorl method), hydroxyproline (Bonnet and Kopp, 1984), nitrate (brucine method), nitrite (ISO 2918) and NaCl content (Carpentier-Volhard method).

Fatty acid composition was carried out on total of fat extract according Bligh and Dyer (1959) after methyl-derivatization (Morrison and Smith, 1964). Gas chromatographic analysis was performed with a Perkin-Elmer Auto syst-X.L equipped with an Omegawax 320 column. Results are presented as g/100g fatty acids (% by weight) of oleic acid, saturated, monounsaturated and polyunsaturated acids. Nutritional quality is described by the polyunsaturated:saturated ratio and ω_6:ω_3 ratio.

Surface colour of the pieces was measured using a reflectance spectrophotometer (Minolta CM-2002). Colour coordinates were determined in the CIE-LAB system and the results were expressed as lightness (L*), redness (a*) and yellowness (b*).

Instrumental Texture Profile Analysis (TPA) (Breene, 1975) was used to evaluate the instrumental texture. All the measurements were made using Texture Analyzer TA-XT2 (Stable Micro Systems Ltd.) with cylindrical probe. A uniaxial compression test was carried out using a cross-head speed of 1 mm/min and the level of compression was 50% of sample thickness. From the TPA curves hardness, springiness, chewiness and cohesiveness were the parameters obtained.

Sensory evaluation was carried out on "Cecina" slices by an trained 8-member sensory panel. The parameters studied were: external appearance (colour homogeneity, colour intensity, marbling, intermuscular fat and yellowness), odour intensity, texture (hardness, chewiness, juiciness, and pastiness) and flavour (flavour intensity and after taste). The sensory attributes were scored using 5-points hedonic scales, 5 denoted extremely high and 1 denoted extremely low.

Statistical analysis of data was carried out by one-way analysis of variance, and means were separated by Tukey honest significant difference test using at 5% level (Statistica software package).

Results and discussion

The chemical composition and physicochemical parameters of the three types of "Cecina" are shown in Table 1. No differences ($p>0.05$) were observed in pH and a_w. The values of a_w (0.882-0.903) are characteristic of an intermediate moisture meat product, which give microbial stability and, hence, help to preserve meat. In relation to the proximate composition, except for moisture content, "contra" was significantly different ($p<0.05$) from "babilla" and "tapa". The results obtained for protein, fat, ash and hydroxyproline contents in "babilla" and "tapa" were basically in line with data published by Gutierrez et al. (1998) for "tapa". The small differences ($p<0.05$) existing in moisture content between "babilla" (52.4%) and the other pieces, "tapa" and contra", could be attributed to its lower size since for the same ripening time the water loss is higher. Although "Cecina de León" is characterized by a high protein and low fat content, "contra" presented a high ($p<0.05$) fat content (28.6% dry matter) resulting in a low ($p<0.05$) protein, carbohydrate and ash contents. Differences in histochemical composition between muscles could explain the differences found in fat content between retail cuts used (Hunt and Hedrick, 1977; Renerre, 1984; Kirchoefer et al., 2002). In this sense, some muscles included in "contra" (M. *gluteobiceps* and *semitendinosus*, overall its inner part) are characterized by a higher percentage of red fibbers, with greater fat contents, than those included in "tapa" (M. s*emimembranosus* and *gracilis*).

Table 1.-Chemical composition and physicochemical parameters (mean ± S.D.) of "babilla" (n=10), "tapa" (n=10) and "contra" (n=10).

Parameters	***"Babilla"***	***"Tapa"***	***"Contra"***
pH	^{a} 5.9 ± 0.28	^{a} 5.8 ± 01	^{a} 5.9 ± 0.12
a_w	^{a} 0.890 ± 0.021	^{a} 0.903 ± 0.011	^{a} 0.882 ± 0.020
Moisture (%)	^{a} 52.4 ± 3.8	^{b} 56.0 ± 2.0	^{b} 57.0 ± 3.0
Protein (% DM)	^{a} 72.8 ± 7.8	^{a} 70.8 ± 3.6	^{b} 60.0 ± 8.4
Fat (% DM)	^{a} 13.2 ± 3.0	^{a} 12.56 ± 3.0	^{b} 28.9 ± 9.0
Ash (% DM)	^{b} 14.8 ± 1.26	^{b} 16.6 ± 1.5	^{a} 12.26 ± 1.5
Carbohydrate (% DM)	^{b} 0.53 ± 0.31	^{b} 0.92 ± 0.15	^{a} 0.13 ± 0.05
Hydroxyproline (% DM)	^{a} 0.45 ± 0.07	^{a} 0.4 ± 0.07	^{b} 0.65 ± 0.25
Oleic acid (%)	^{a} 35.9 ± 2.6	^{b} 40.6 ± 2.2	^{b} 42.9 ± 2.0
Saturated (%)	^{b} 42.5 ± 2.6	^{ab} 41.5 ± 3.2	^{a} 38.7 ± 2.1
Monounsaturated (%)	^{a} 39.1 ± 2.7	^{b} 47.8 ± 3.7	^{b} 50.0 ± 3.1
Polyunsaturated (%)	^{a} 6.3 ± 2.4	^{a} 4.8 ± 0.9	^{a} 4.4 ± 1.0
Unsaturated (%)	^{a} 45.4 ± 2.0	^{b} 53.0 ± 3.1	^{b} 54.4 ± 3.1
Polyunsaturated/Saturated	^{a} 0.15 ± 0.06	^{a} 0.11 ± 0.02	^{a} 0.11 ± 0.02
ω_3	^{b} 1.8 ± 1.4	^{a} 0.6 ± 0.2	^{a} 1.0 ± 0.3
ω_6	^{a} 4.3 ± 1.3	^{a} 3.3 ± 1.0	^{a} 3.4 ± 0.9
ω_6/ω_3	^{a} 3.5 ± 2.1	^{b} 6.2 ± 2.5	^{a} 3.5 ± 1.1
NaCl (% DM)	^{a} 8.6 ± 1.7	^{b} 13.0 ± 1.2	^{a} 9.6 ± 0.9
Nitrate (ppm DM)	^{a} 120.0 ± 46	^{b} 163.0 ± 40	^{b} 160.7 ± 40
Nitrite (ppm DM)	n.d.	0.9 ^{a} ± 0.1	2.8 ^{b} ± 0.8

$^{a, b, c}$ Means with different letters indicate significant differences (Tukey test: $p<0.05$).
DM: dry matter; n.d.: not detected.

Regarding fatty acid composition, "babilla" showed a lower ($p<0.05$) content than "tapa" and "contra" in oleic acid (35.9% vs. 40.6 and 42.9% respectively), and in consequence in monounsaturated and unsaturated fatty acids. "Babilla" also presented the higher polyunsaturated fatty acids contents, however no differences ($p>0.05$) between pieces were found for this parameter. The amount of saturated fatty acids was significantly different ($p<0.05$) in "babilla" and "contra". The ratio between polyunsaturated and saturated fatty acids was below to that recommended for human diet (>0.4). The highest ($p<0.05$) ω_6/ω_3 was found in "tapa" (6.2), "babilla" and "contra" presented a more favourable balance (3.5) between ω_6 and ω_3 polyunsaturated acid. A dietary ratio of 4 or 5:1 for ω_6/ω_3 polyunsaturated acids is desirable. Higher ratios are less desirable. The proportion is especially important in relation to the incidence of cardiovascular disease (Warriss, 2000).

Concerning the curing agents, the salt content was higher ($p<0.05$) in "tapa", significant differences ($p<0.05$) were also found in nitrite and nitrate content. These values are under their legally established limits (Directive 2001/5/CEE).

Results of colour measurement are shown in Table 2. No differences ($p>0.05$) in L* value were detected among the three pieces. "Babilla" had lower ($p<0.05$) a* and b*, as compared to those of "tapa" and "contra".

Table 2.- Values (mean ± S.D.) of colour parameters in "Cecina".

CIE –LAB coordinates	*"Babilla"*	*"Tapa"*	*"Contra"*
L* (lightness)	^{a} 30.0 ± 2.2	^{a} 29.3 ± 1.8	^{a} 31.7 ± 2.6
a* (redness)	^{a} 7.8 ± 2.5	^{b} 11.6 ± 3.0	^{b} 10.0 ± 2.3
b* (yellowness)	^{a} 1.9 ± 1.4	^{b} 5.7 ± 2.4	^{b} 5.8 ± 2.0

$^{a, b, c}$ Means with different letters indicate significant differences (Tukey test: $p<0.05$).

The values of texture descriptors are shown in Figure 1. No differences ($p>0.05$) were found in cohesiveness and springiness among the three type of "Cecina". On the contrary, "contra" exhibited higher ($p<0.05$) values, both in hardness and chewiness than "babilla" and "tapa". The hydroxyproline content has been previously fitted as indicator for hardness (Rodriguez-Lazaro *et al*., 2001). In this sense, "contra" was the piece that presented a higher ($p<0.05$) hydroxyproline content, this is explained by the higher hydroxyproline content in M. *semitendinosus* (included in "contra") than M. *semimembranosus* (included in "tapa") (Pedersen *et. al*., 1996).

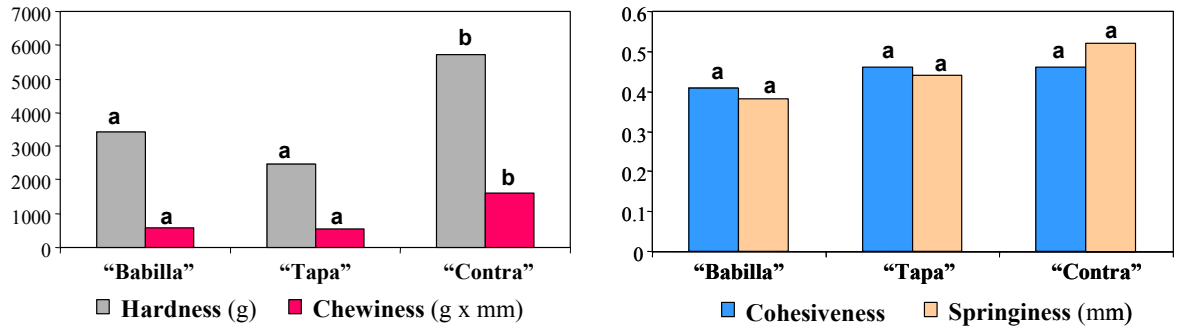

Figure 1.- Mean values of Instrumental Texture Profile Analysis (TPA). (^{abc} Columns with different letters indicate significant differences (Tukey test: $p<0.05$).

Table 3.- Results (mean ± S.D.) of sensorial analysis of the three types of "Cecina".

	"Babilla"	*"Tapa"*	*"Contra"*
Colour homogeneity	^{a} 3.7 ± 0.2	^{a} 3.6 ± 0.4	^{a} 3.4 ± 0.9
Colour intensity	^{a} 3.5 ± 0.5	^{a} 3.5 ± 0.4	^{a} 3.8 ± 0.5
Marbling	^{a} 2.9 ± 0.4	^{a} 2.9 ± 0.8	^{a} 2.5 ± 1.3
Intermuscular fat	^{ab} 3.0 ± 0.5	^{a} 2.2 ± 0.4	^{b} 3.7 ± 1.2
Yellowness	^{a} 1.3 ± 0.4	^{b} 1.8 ± 0.2	^{a} 1.3 ± 0.4
Odour intensity	^{a} 3.0 ± 0.3	^{a} 3.0 ± 0.3	^{a} 3.1 ± 0.6
Hardness	^{a} 2.4 ± 0.8	^{a} 2.3 ± 0.5	^{b} 3.0 ± 0.9
Chewiness	^{a} 2.0 ± 0.3	^{a} 2.1 ± 0.3	^{b} 3.5 ± 0.5
Juiciness	^{a} 3.1 ± 0.2	^{b} 3.5 ± 0.4	^{a} 2.8 ± 0.4
Pastiness	^{a} 1.2 ± 0.1	^{a} 1.2 ± 0.1	^{b} 2.4 ± 0.4
Flavour intensity	^{a} 3.0 ± 0.4	^{a} 3.1 ± 0.3	^{a} 3.1 ± 0.6
After taste	^{b} 3.8 ± 0.1	^{a} 2.6 ± 0.5	^{b} 3.1 ± 0.4

^{a,b,c} Means with different letters indicate significant differences (Tukey test: $p<0.05$).

Results of sensory analysis of "Cecina" are presented in Table 3. No differences ($p>0.05$) were found in colour (homogeneity and intensity), odour intensity and flavour intensity. Concerning fat, marbling was similar ($p>0.05$) between the evaluated pieces, however the presence of intermuscular fat was higher ($p<0.05$) in "contra" than "tapa", which presented the highest yellowness scores ($p<0.05$). On the other hand, the highest values ($p<0.05$) for hardness, pastiness and chewiness obtained in "contra" confirm the instrumental texture results. Finally, the judges considered that "tapa" was more juicy ($p<0.05$) than "babilla" and "contra" although its aftertaste was lower ($p<0.05$).

Conclusions

"Babilla" and "tapa" seem to be the most similar between the types of "Cecina" studied. Both pieces of "Cecina" presented a higher protein content and a lower fat content than "contra". However, "contra" showed the highest acid oleic percentage and a good favourable balance between ω_6 /ω_3 polyunsaturated acids. By the other hand, instrumental Texture Profile Analysis (TPA), as well as sensory evaluation, indicated that "contra" was the hardest piece of "Cecina" studied, but also it had higher chewiness than the other.
Further research is needed for a better understanding of the relationship between anatomical origin of raw meat and final meat product.

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EFFECT OF DIFFERENT CONCENTRATIONS OF NATURAL COLORANTS ON THE COLOUR OF FRESH PORK SAUSAGES PACKAGED IN MODIFIED ATMOSPHERE

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Background

Colour is the most deleterious factor as regards the appearance of meat and meat products during storage. It is also the most influential on the consumer purchasing decision. The attractive bright cherry red colour of freshly cut or comminuted meat is used by consumers as an indication of freshness. The preferences of the consumers for natural colorants has been increasing during these last years, because "natural" is often associated with an image of healthy and good quality. Consumers tend to perceive synthetic colorants as undesirable and harmful, and some are considered to be responsible for allergic and intolerance reaction (Blenford, 1995). Stability has been a problem with the use of natural colorants which are often very sensitive to heat, light, acidity, air or water activity changes (Attoe and von Elbe, 1981; Carnevale et al., 1980; Von Elbe et al., 1974). The combined use of colorants and modified atmosphere packaging for meat represents a realistic and attractive strategy to increase the shelf life of fresh meat and fresh meat products. Monascus species produce red colorants through solid state fermentation and have been used as a general food colorant and medical agent for centuries (Went, 1895). Recently, these pigments were tested for their ability to colour different foods, including meat products (Fink-Gremmels and Leistner, 1989). The red beet root (*Beta vulgaris*) is a rich source of red pigments known as betalains (Mabry and Dreiding, 1968).

Objectives

To study the effects of natural pigments of mould *Monascus purpureus* and red beet root juice, compared to betanin (E-162), on the shelf-life of fresh pork sausages packaged in modified atmosphere, in order to select the most appropriate natural colorant for improving their quality.

Materials and methods

Preparation of samples. Four pork forelegs were obtained at 48 h post slaughter from a local supplier (MARBE, Zaragoza, Spain), trimmed of external fat, and ground using an industrial grinder machine. Minced meat was divided in nine batches, which were mixed with NaCl (to a final concentration of 2%) and with either: 1) Control (no colorants), 2) *Monascus purpureus* (0.05%), 3) *Monascus purpureus* (0.1%), 4) *Monascus purpureus* (0.2%), 5) Red beet root juice (0.5 ml / kg meat) , 6) Red beet root juice (1 ml / kg meat), 7) E-162 (0.03%), 8) E-162 (0.05%), 9) E-162 (0.07%). The fresh sausages were stuffed into collagen casings, Colfan F (Viscofan S.A., Caseda, Spain), placed on polypropylene trays, introduced in a pouch made of a polyethylene and polyamide and filled with 80% O2 + 20% CO2 gas mixture. Sausages were stored for 16 days at 2 ± 1°C in the dark.

Red beet juice preparation. Fresh red beet roots (*Beta vulgaris*) were purchased from a commercial store. Samples were washed, dried and cut into cubes of about 1cm x 1cm, which were boiled for 5 min for blanching (Han et al, 1998). After rapid cooling, beet juice was extracted with a standard kitchen food processor. The crude juice was boiled at 100ºC for 1 min. After rapid cooling, it was filtered stepwise (MN 640w, Machinery Nagel GmbH & Co. KG, Düren, Alemania). The clear beet juice was stored at refrigeration until use.

Colour measurement. Meat colour was measured at the sausage surface using a reflectance spectrophotometer (Minolta CM-2002; Osaka, Japan), 30 min after of opening the packing. CIE L*, a*, b* (CIE, 1978) parameters were recorded. The h* value and C* were calculated $h^* = \tan^{-1}(b^*/a^*)$ and $C^* = ((a^*)^2+(b^{*2}))^{0.5}$. The spectra curves were determined over the range of 400-700 nm at 10 nm intervals with Minolta CM-2002. Each value was the mean of 30 determinations.

Statistical analyses. The significance of differences among samples at each day of storage was determined by analysis of variance using the least square difference method of the General Linear Model procedure of

SPSS (SPSS 11.5, 2002). Differences were considered significant at the $p<0.05$ level. Data were also analysed using canonical discriminant analysis (CDA) multivariate statistical method of SPSS (SPSS 11.5, 2002).

Results and discussion

The evolution of different colour parameters is shown in Table 1. Lightness. The use of colorants caused a decrease of L* values in all treatments ($p<0.05$) throughout the whole study. All samples with colorants added were darker than control fresh pork sausages, the higher the concentration the lower the lightness. Bloukas et al. (1999) found that betanin level significantly affected lightness, the higher the betanin level the lower the L* values. Stuempel (1997) also reported lower L* values for frankfurter-type sausages with higher betanin levels; Pipek et al. (1996) found that Monascus extract caused a decrease of brightness in sausages and frankfurters.

Redness. Samples with natural colorant added had higher a* values ($p<0.05$) than control sausages, the higher the concentration the higher the redness. Sayas-Barberá et al. (1987) also found a relationship with a* values and Monascus concentrations. Pipek et al. (1996) found that Monascus extract caused an increase of a* values. The behaviour of red beet root might be explained by betanin degradation by air and light. Bloukas et al. (1999) found that redness of frankfurters steadily increased with the betanin level; similar results were reported by Stuempel (1997).

Yellowness. Samples did not present variations in b* values along the 16 days of storage ($p>0.05$). Neither Klettner (1993), Pipek et al. (1996) nor Sayas-Barberá et al. (1987) found relevant variations in b* values. However, Bloukas (1999) found that use of betanin in frankfurters increased b* values.

a*/b* ratio. Changes of this ratio were very similar to those of a* values.

Chroma. All samples showed a slight increase of C* values at 4th day of storage; after that values decreased ($p<0.05$) along all the storage period. E-162 (0.07%) showed the higher chroma values, near 19, during the first 8 days after that they suffered a very significant decrease. Only Monascus 0.05% presented lower values throughout the experiment ($p<0.05$). Decrease or increase in C* has been associated to increase or decrease in h*, respectively. The typical pink-red colour of fresh pork meat minced is related to low h* and high C* values this colour changed to oxidized brown-grey meat surface (high h* and low C* values).

Hue. As the colour attribute hue increased, the shade of colour changed from red to brown, a gradual rise of hue angle indicated increasing metmyoglobin during storage (Isdell et al., 1999). Changes in hue angles confirmed the results obtained by measuring only a* values. Sayas-Barberá et al. (1987) found that the hue value decreased significantly by using above 40 ppm of Monascus.

Reflectance spectra of control fresh pork sausages and meat with *Monascus purpureus* (0.05%), E-162 (0.05%) and red beet root (1ml) are shown in Fig. 1. The reflectance spectrum of meat with red beet root was very similar to that of the fresh sausage used as control. The use of E-162 gave rise to a spectrum similar to the control, but was lower between 440 and 540 nm and from 590 nm to 700 nm. The spectrum of Monascus was the most different, presenting the lowest reflectance values. Palombo and Wijngaards (1990) pointed out that the shift of the spectrum towards higher reflectance values was due to a remarkable increase of L*, a decrease of h* and a decrease of C* values.

Canonical discriminant analysis is shown in Fig. 2. The two fist canonical functions accounted for 96.2% of variability of fresh pork sausages, therefore, they are a valuable tool for discriminating the different treatments. The most different treatments were Monascus (negative region) and control (positive region); red beet root at different concentrations had an intermediate behaviour between Monascus and the control samples.

Conclusions

The use of natural food colorant improved some colour characteristics of the fresh pork sausages. All samples with *Monascus purpureus*, betanin (E-162) and red beet root presented lower L*, b* and h* values

than control sausages, and higher a*, a*/b* and C* values than control. If red colour is used by consumers as freshness indicator and as willingness of purchasing, the use of natural red colorant enhanced the shelf life of this product. Reflectance spectrum of sausages with red beet root was very similar to control; so this colorant appeared to be most suitable for improving the natural colour of fresh pork sausage.

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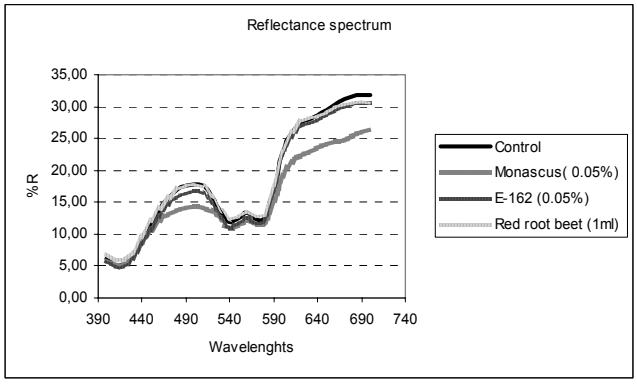

Fig.1. Reflectance spectra of control fresh pork sausages and meat with *Monascus purpureus* (0.05%), E-162 (0.05%) and red beet root (1ml).

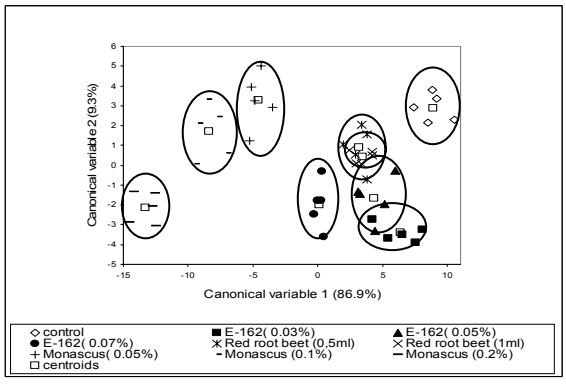

Fig2. Canonical discriminant analysis of control fresh sausages and meat with different concentrations of Monascus purpureus (0.05%, 0.1%, 0.2%), E-162 (0.03%, 0.05%, 0.07%) and red beet root (0.5; 1ml).

Table 1. Evolution of different colour parameters of fresh pork sausages packaged in modified atmosphere with different concentrations of natural colourants.

		Days of storage				
	Treatments	**0**	**4**	**8**	**12**	**16**
L*	Control	43.33aw	43.68aw	44.27aw	45.13avw	47.86av
	E-162 (0.03%)	38.10cw	38.15cw	38.44cw	39.60cvw	41.93cv
	E-162 (0.05%)	38.83cw	37.55cw	38.85cw	40.26bcv	41.21cv
	E-162 (0.07%)	38.24cw	36.83cw	37.61cw	39.74cv	40.00cv
	Red root beet (0,5ml)	41.00bw	40.59bw	41.83abw	41.45bw	46.15av
	Red root beet 1ml	40.73bw	40.18bw	41.13bw	41.28bw	44.94abv
	Monascus (0.05%)	40.73bw	41.87abw	43.15av	42.83bvw	44.63bv
	Monascus (0.1%)	39.6bcw	41.15abvw	41.12bvw	40.01bcw	42.58cv
	Monascus (0.2%)	38.40cvw	39.13bv	39.45cv	37.9dw	38.25dvw
a*	Control	9.31dv	10.08ev	9.32ev	6.06dw	1.25ex
	E-162 (0.03%)	10.80cv	11.25dv	10.11dv	8.73cw	3.50cx
	E-162 (0.05%)	13.01bv	13.30cv	12.18bw	11.51bw	6.08bx
	E-162 (0.07%)	14.00abv	14.61bv	14.57av	11.28bw	6.89bx
	Red root beet (0,5ml)	10.91cv	11.42dv	9.97dew	6.88dx	1.67dey
	Red root beet 1ml	11.15cv	11.58dv	10.04dw	8.44cx	2.33dy
	Monascus (0.05%)	10.08cdw	11.60dv	11.18cv	8.61cx	2.49dy
	Monascus (0.1%)	12.63bv	13.03cv	12.58bv	10.70bx	5.68by
	Monascus (0.2%)	14.89av	15.60av	14.94av	14.02aw	12.40ax
b*	Control	13.21av	13.50av	13.40av	13.54av	13.20av
	E-162 (0.03%)	13.00av	13.20av	13.30av	13.30av	12.60av
	E-162 (0.05%)	12.74av	12.97av	13.01av	13.10av	12.48av
	E-162 (0.07%)	12.32av	12.40av	12.39v	12.34av	12.30av
	Red root beet (0,5ml)	12.43av	12.60av	12.67av	12.58av	12.48av
	Red root beet 1ml	12.53av	12.70av	12.75av	12.50av	12.60av
	Monascus (0.05%)	11.24bv	11.35bv	11.40bv	11.22bv	11.47bv
	Monascus (0.1%)	10.60bcv	10.92bcv	10.62bcv	10.98bcv	10.7bcv
	Monascus (0.2%)	9.72cv	10.11cv	9.77cv	10.10cv	10.04cv
a*/b*	Control	0.70fv	0.75fv	0.70dv	0.45fw	0.09fx
	E-162 (0.03%)	0.83ev	0.85ev	0.76dw	0.66dx	0.28dy
	E-162 (0.05%)	1.02cv	1.03cv	0.94cv	0.88bw	0.49cx
	E-162 (0.07%)	1.14bv	1.18bv	1.19bv	0.92bx	0.57by
	Red root beet (0,5ml)	0.88dv	0.91dv	0.79dx	0.55ey	0.13ez
	Red root beet 1ml	0.89dv	0.91dv	0.79dw	0.68dx	0.18ey
	Monascus (0.05%)	0.90dw	1.02cv	0.98bx	0.77cy	0.23dz
	Monascus (0.1%)	1.19bv	1.19bv	1.19bv	0.97bx	0.53cy
	Monascus (0.2%)	1.53av	1.54av	1.53av	1.42ax	1.23ay
C*	Control	16.16bv	16.85bcv	16.32cv	14.83bw	13.26bx
	E-162 (0.03%)	16.90bv	17.30abv	16.70cv	15.91abw	13.08bx
	E-162 (0.05%)	18.21av	18.57av	17.82bvw	17.44aw	13.88bx
	E-162 (0.07%)	18.65av	19.16av	19.05av	16.62aw	13.86bx
	Red root beet (0,5ml)	16.54bv	17.01bv	16.13cv	14.34bcw	12.59bcx
	Red root beet 1ml	16.77bv	17.19bv	16.23cv	15.08bw	12.81bcx
	Monascus (0.05%)	15.10cvw	16.22cv	15.96cv	14.14cw	11.74cx
	Monascus (0.1%)	16.49bv	17.00bv	16.46cv	15.33bw	11.98cx
	Monascus (0.2%)	17.78av	18.59av	17.85bv	17.53av	15.96ax
H*	Control	54.82ax	53.26ax	55.18ax	65.89aw	84.60av
	E-162 (0.03%)	50.28bx	49.57bx	52.77abx	56.72cw	74.49cv
	E-162 (0.05%)	44.39cwx	44.28cx	46.89cw	48.70ew	64.02dv
	E-162 (0.07%)	41.35dx	40.32dx	40.10dx	47.29ew	60.16ev
	Red root beet (0,5ml)	48.71by	47.80by	51.79bx	61.33bw	82.36av
	Red root beet 1ml	48.33by	47.65by	51.76bx	55.96cw	79.53bv
	Monascus (0.05%)	48.12bx	44.38cy	45.57cy	52.48dw	77.76bv
	Monascus (0.1%)	40.01dx	39.96dx	40.16ex	45.75ew	63.27dv
	Monascus (0.2%)	33.14ew	32.95ew	33.19fw	35.20fw	39.01fv

pH, WATER ACTIVITY, AND PROXIMATE COMPOSITION OF *MORCILLA DE LEÓN*, A TRADITIONAL EUROPEAN BLOOD SAUSAGE

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Background

Among the diverse types of meat products in the world, there are some typical products which are elaborated with blood; these are named as "blood sausages". These sausages have rarely been studied, being found some bibliographic references in Germany (Souci, *et al*., 1989; Stiebing, 1990), Spain (Antiduelo, 2002; Santos *et al*., 2003) and Portugal (Roseiro *et al*., 1998) and some countries from Latin American (Bunger *et al*., 1992, Adesiyun *et al*., 1996). Blood sausages are made (besides blood) with ingredients such as fat, offal, onion, cereals, spices, etc., existing a great diversity of ingredients as well as ways of processing and preparation. The *Morcilla de León* is a typical product from Castilla and León (Spain), being onion (65%–75%), pork and/or beef fat (10%–20%), pork and/or beef blood (10%–20%), rice (2%–5%), salt (1%–1.5%) and spices the main ingredients in its composition (Antiduelo, 2002). Process usually consists on precooking the mixed of ingredients, which are then stuffed into a pork casing and cooked in hot water (80-100ºC). Finally, *Morcilla de León* is consumed fried or boiled.

Objectives

This study is aimed to contribute to *Morcilla de León* typification, a typical regional Spanish sausage, by studying its proximate composition and comparing results from this blood sausage with other results from different blood sausages in the World.

Materials and methods

Samples of blood sausages were purchased from the retail market coming from nine different meat industries which were located in León City and other smaller towns nearby. Once acquired, the samples were immediately transported to laboratory in refrigerated conditions. There, samples were homogenised and the following analysis were accomplished. pH was measured in fresh sample by duplicating according to the official method for meats (Presidencia del Gobierno, 1979). Water activity (a_w) was measured by a dewdrop point method with an Aqualab CX-2 (Decagon Devices, Washington, USA). Moisture was determined according to ISO (1973). Fat content of the dehydrated sample was measured by extraction with petroleum ether according to AOAC (1999a). Total protein was determined by the Kjeldahl method according to AOAC (1999b), by using 6.25 as conversion factor. Ash content was measured by incineration sample at 550ºC, according to Presidencia del Gobierno (1979). Finally, digestible carbohydrates extraction was carried out with 52% perchloric acid following the Presidencia del Gobierno (1982) method and quantification was made with the method described by Dubois *et al*. (1956). Relative analyses on composition were determined by duplicate and data were expressed as percentage on fresh and dry matter.

Results and discussion

pH and a_w values for *Morcilla de León* are shown at table 1, in which it is indicated that the pH averages c.a. 6 and the a_w averages 0.97. These parameters, which depend mainly on used ingredients in the sausage, i.e. onion, blood, fat and salt, were similar to those found by Santos *et al.* (2003) in the '*Morcilla de Burgos'*, another typical blood sausage from Spain made mainly with onion, rice, blood and fat. Due to the high pH and a_w of the sausage and the absence of preservatives in the formula, *Morcilla de León* is susceptible to early spoilage, its shelf-life depends on the initial microbial population of the mixture, heat treatment intensity in the boiling stage, handling conditions after boiling, and storage temperature. Anyway, during chilled storage a pH decrease (c.a. 0.5) has normally been observed which could be attributed to microbial growth (non published data).

Proximate composition of *Morcilla de León* is shown at tables 2 and 3. Regarding to moisture content, it was also similar to that of *Morcilla de Burgos* (Santos *et al*., 2003) but 11% lower than blood sausage from Chile (Bunger *et al*., 1992) and 21% and 25% higher than the moisture of *Morcela de Assar* from Portugal (Roseiro *et al*., 1998) and German '*Blutwurst'* (Souci *et al*., 1990), respectively. The reason for this variability could be explained by the differences in the types and amounts of ingredients used in the formulation of each kind of blood sausages. Furthermore, fat is probably the most variable proximate component in meat products. The fat content on dry matter found in different blood sausages has had a range between 28% of Morcilla de Burgos to 70% of Blutwurst (Souci, *et al.*, 1990, Bunger *et al.*, 1992, Roseiro *et al*., 1998, Santos *et al.*, 2003,), and the fat content on dry matter of *Morcilla de León* was into an intermediate place. In the same way, protein percentage on dry matter of *Morcilla de León* was slightly higher than those found in *Morcilla de Burgos,* and lower than those of other blood sausages.
Digestible carbohydrates in the *Morcilla de León*– fraction constituted basically by starch and soluble sugars – were 27% of the dry matter. This percentage was rather lower than in *Morcilla de Burgos* –51% on dry matter– because more rice (20% to 35% of rice) is added to make the last sausage (Santos *et al*., 2003) while the amount of rice or bread that is added to the *Morcilla de León* is about up to 10% (Antiduelo, 2002). The amount of cereals or other vegetables used for making other blood sausages such as Blutwurst, Chilean blood sausage or *Morcela de Assar* is even less and due to it its results of digestible carbohydrate. The presence of more than 1% of fiber in the *Morcilla de León* as well as in other blood sausage (Bunger et al., 1992; Santos et al., 2003) is also due to the addition of vegetables as ingredients, such as the onion, which, as it was said before, is the principal ingredient of *Morcilla de León* (c.a. 70%). Fiber content of cooked onion is approximately 1.5% (USDA, 2003). Finally, ash content of *Morcilla de León* was in the higher part of the range observed for blood sausages (4.3% a 8.4% on dry matter) (Roseiro *et al*., 1998, Santos *et al.*, 2003, Bunger *et al.*, 1992, Souci, *et al.*, 1990).
Table 4 contains the correlations between moisture, fat, ash and protein contents of *Morcilla de León* expressed as dry matter. Only the correlation digestible carbohydrates *vs.* fat –the two most abundant components– was statistically significant ($p<0.05$).

Conclusions

'Morcilla de León' is a cooked product subject to easy microbial spoilage which needs chilled storage. It represents a nutritional source of fat, carbohydrates, fiber and protein –mainly from blood–; nonetheless it has less protein than meat and most of meat products.

Acknowledgements

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Table 1. Mean, standard deviation (S.D.), maximum and minimum values of pH and a_w of *Morcilla de León*.

	Mean	S.D.	Minimum	Maximum
pH	6.1	0.4	6.0	7.1
a_w	0.972	0.003	0.969	0.977

Table 2. Proximate composition of *Morcilla de León* expressed as percentage of fresh matter.

	Mean	S.D.	Minimum	Maximum
Moisture	66.2	4.8	59.8	75.0
Fat	14.2	3.0	9.0	17.2
Total protein	5.7	0.8	4.4	7.0
Ash	1.9	0.2	1.7	2.1
Digestible carbohydrates[#]	9.0	1.8	6.4	10.8
Fibre[*]	3.0	---	---	---

[#] Expressed as % of glucose, [*] Estimated by difference.

Table 3. Proximate composition of *Morcilla de León* expressed as percentage of dry matter.

	Mean	S.D.	Minimum	Maximum
Fat	42.1	6.6	32.1	51.3
Total protein	17.1	2.8	13.0	20.8
Ash	6.6	1.4	4.9	8.6
Digestible carbohydrates[#]	27.0	6.1	20.1	38.8
Fibre*	7.2	---	---	---

[#] Expressed as % of glucose, [*]Estimated by difference.

Table 4. Correlations between the parameters of the proximate composition of *Morcilla de León*, expressed in terms of dry matter.

	Moisture	Fat	Ash	Total protein	Digestible carbohydrates
Moisture	1.00	-0.08	0.05	0.63	0.41
Fat	---	1.00	0.20	-0.00	**-0.74**
Ash	---	---	1.00	-0.27	-0.07
Total protein	---	---	---	1.00	-0.14
Digestible carbohydrates	---	---	---	---	1.00

Values in bold were significant ($p<0.05$)

THE INFLUENCE OF WINE COMPONENTS ON THE TENDERNESS AND HISTOLOGICAL STRUCTURE OF COOKED MEAT

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Background

Marinating in wine have long been used as a mean of reducing toughness and consequently to improve tenderness of beef muscles. In the case of typical German cuisine "Sauer Brauten", beef round meat was soaked in red wine for 2-3days and subsequently was cooked. Some authors reported that the tenderizing effect of wine was related to various components of wine (organic acids, sugar, polyphenol and alcohol [1), 2), 3), 4)]. However more studies are necessary to examine the relationship between the components of wine and meat tenderness.

Keywords: Wine, marinating, cooked meat tenderness, histological structure

Objectives

The aim of this study was to find the effect of various components of red and white wine on the meat tenderness and histological structure.

Materials and methods

Beef round meat was obtained from commercial source. After the fat and tendon (perimycium) were removed, the meat was cut into pieces weighing about 150g (2cm thick). Each piece of meat was put into plastic bag(Asahi Kasei H N type) and then soaked in each solution(water, white wine, red wine, 0-50mg Tannin, 0-0.6%tartarir acid, 0-2%glucose, 0-20%ethylalcohol) for 5 hours. These samples were heated at 100□ for 30min in the water bath. Thereafter five 1.27cm diameter cores, taken parallel to the muscle fiber, were removed from one piece per sample. Cores were sheared with Warner-Blatzler Meat Shear Model 2000. Shear force Values were recorded as kg force/1.27 cm sample. Samples for histological observation of meat structure were frozen in the freezer and cut into thin vertical sections. Observations were performed using an optical microscope (Nikon optiphoto).

Results and discussion

Shear force value of cooked meat

Figure 1 shows the mean and individual shear force values obtained for cooked meat marinating in water, red wine and white wine. There were no significant differences between red wine, white wine and water (control).

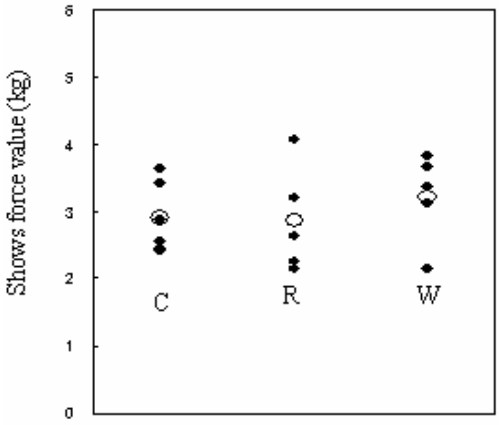

Fig.1

Figure 1 shows the shear force value of cooked meat marinated in C: water, R: red wine, W: white wine

Figure 2-a,b,c,d showed the shear value of cooked meat marinating in tannin, tartaric acid, glucose and ethyl alcohol, respectively.
As, shown in Figure 2-a, the concentration of tannin increased from 0 to 50 mg%, the shear force value of cooked meat increased from 2.4 kg to 3.9kg (correlation coefficient=0.67). There was a similar trend for the increase of glucose concentration (Figure 2-c). Whereas Figure 2-b indicated that there was the excellent correlation (-0.95) between decreased tenderness and increased tartaric acid concentration from 0 to 0.6%. A similar trend was observed in increasing of ethyl alcohol concentration (Figure 2-d).

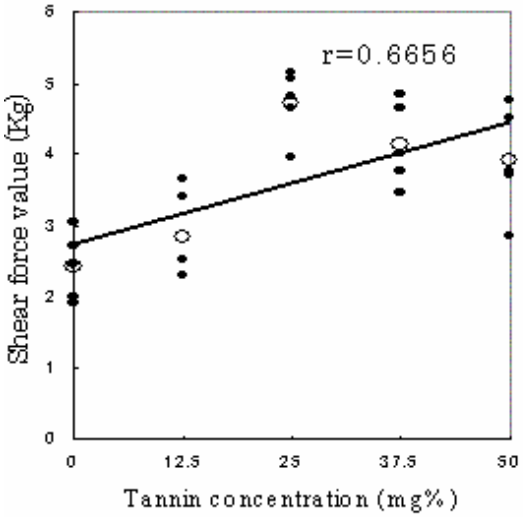

Fig.2-a

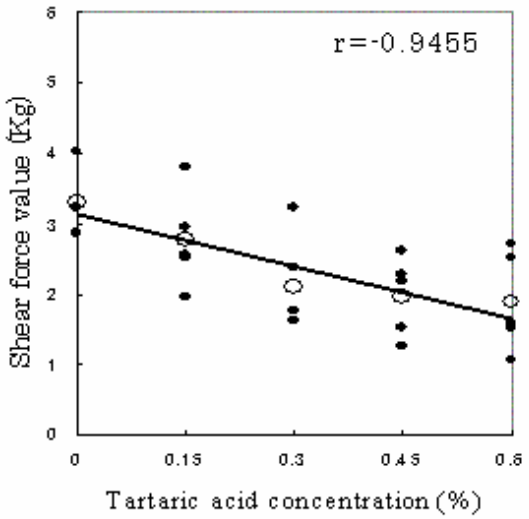

Fig.2-b

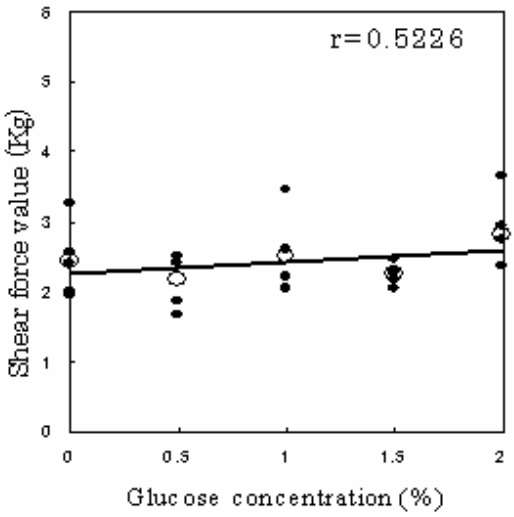

Fig.2-c

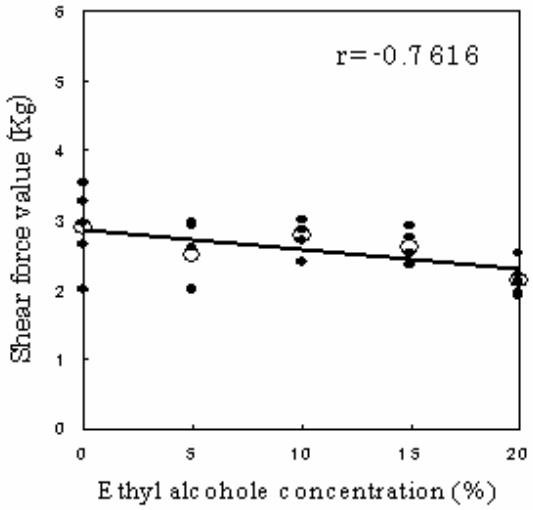

Fig.2-d

Figure 2 compares the shear force value of cooked meat marinated in (a) tannin, (b) tartaric acid, (c) glucose, (d) ethyl alcohol.

Structure of cooked meat

Figure 3-a,b,c,d,e showed the structure of marinated meat in water, tannin, tartaric acid, glucose and ethyl alcohol. The marinating in tannin leads to the remarkable shrinking of muscle fiber, simultaneously, that appearance also observed for marinating in glucose. Whereas in the case of marinating in tartaric acid and in ethyl alcohol no shrinkage occurred, but appeared horizontal cracks in muscle fiber structure.

Fig.3-a Water

Fig.3-b Tannin

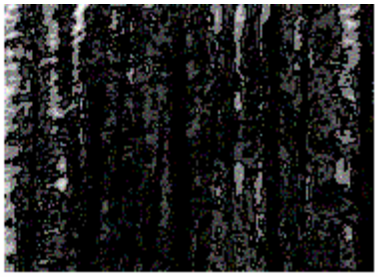

Fig.3-c Tartaric acid

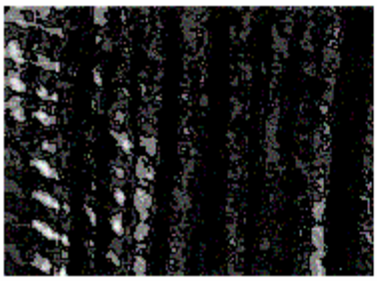

Fig.3-d Glucose

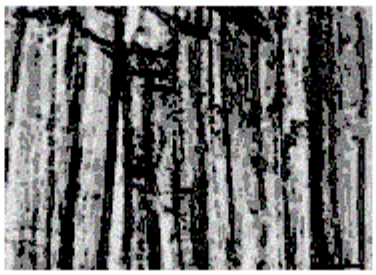

Fig.3-e Ethyl alcohol

Figure 3 showed the microstructure of cooked meat marinated in (a) water, (b) tannin, (c) tartaric acid, (d) glucose, (e) ethyl alcohol.

Conclusions

There were no significant differences in shear force value obtained for cooked beet round meat marinating in red wine, white wine and water. Marinating in tartaric acid was most effective to tenderizing of meat. When the meat soaked in Tannin, the shear force value of cooked meat increased as concentration of tannin increased. This treatment leads to the remarkable shrinking of muscle fiber.
These results indicate that the excellent correlation between increased toughness of cooked meat associated with shrinking of muscle fiber.

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QUALITY OF LOW-FAT MEATBALLS CONTAINING LEGUME FLOURS AS EXTENDERS

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Background

Ground meat is used for the production of a variety of traditional meat products such as meatballs, kebabs and döner in Turkey. The meat is mixed with spices and extenders then formed and cooked. Various extenders and binders such as egg, flours and breadcrumbs are used in ground meat formulations to bind the meat particles. Legumes are economic source of protein and also various minerals. In recent years legumes have been investigated regarding to their potential use in developing functional foods. Inclusion of legumes in the daily diet has many physiological effects in controlling and preventing various metabolic diseases such as mellitus, coronary heart disease and colon cancer (Tharanathan and Mahadevamma, 2003).

Beef sausages extended with common bean flour had higher water holding capacity, lower cooking loss and lower protein content (Dzudie et al. 2002). Modi et al. (2003) investigated the effect of Bengal gram, green gram and black gram flours in meatballs, and gram flour in burgers. Beef patties extended with soy flour and samh flour had lower water activity and moisture content but protein content was not affected by the addition of extenders (Elgasim and Al-Wessali, 2000).

Objectives

The objective of this research was to evaluate the effects of legume flours (blackeye bean, chickpea and lentil) and rusk on proximate composition, fatty acid composition and lipid oxidation of low-fat meatballs.

Materials and methods

Beef as boneless rounds was obtained from the local butcher in Izmir, Turkey. All subcutaneous fat and inter-muscular fat was removed from the muscles and used as the fat source. Lean and fat were ground through a 3 mm plate grinder. Legumes; blackeye bean (*Vigna unguiculata,*23.5% protein, 1.5% fat), chickpea (*Cicer arietinum,* 20.6% protein, 4.5% fat), lentil (*Lens culinary*, 23,5% protein, 1.2% fat) and rusk (12,4 protein, 4.5 fat) used in this study were obtained from a local market. Each legume was soaked (ratio of 1:2 legume to water) for 12 hours and cooked for 1.5 hours in boiling water. Cooked legumes were dried separately in electric oven (100ºC) for two hours and grind in a mill. Similarly rusk was also prepared in the same mill to obtain a fine structure and all above the flours were used as extenders. The minced beef was then mixed with 10% (g/100g) extender, 7% beef fat, 0.3% onion powder, (0.8% spice mix) and 2% salt. Batches of 2 kg of each formulation were mixed with food processor and processed into meatballs (1cm thick and 80 mm diameter) by using a metal shaper.

Moisture and ash content of each meatball were measured by using AOAC procedures. Fat content was determined by chloroform-methanol extraction according to Flynn and Bramblett (1975). Protein content was determined according to Anonymous (1979). Lipids were extracted from 10 g samples with chloroform : methanol (2:1 v/v) (Folch et al.1957) and methylated (Anonymous, 1987). Fatty acid methyl esters (FAME) were analyzed using a gas chromatography (HP5890) fitted with a fused silica capillary column (DB-23, 30 mx 0.25 mm id., 0.25 µm film thickness, J.W.Scientific). The column temperature programmed 100ºC to 220 ºC in 4 ºC/min and 15 min at 220ºC. The injector temperature was set at 220ºC and the detector (FID) temperature was set at 220 ºC. The carrier gas was hydrogen at a flow rate of 1 ml/min. The fatty acids were identified by comparision of the retention times of the sample with those of standards.

Meatball samples were frozen at -18°C for 3 months in polypropylene boxes with lids. On 0 th day and 1st, 2nd and 3rd months of frozen storage , samples were thawed at 4°C and oxidative rancidity of meatballs was determined by thiobarbuturic acid test (TBA) according to Tarladgis et al. (1960). Data were subjected to one-way analysis of ANOVA (Minitab, 2003).

Results and discussion

Mean values for proximate composition of uncooked and cooked meatballs are given in Table 1. For uncooked and cooked samples the moisture, fat and ash contents in the formulations of different extenders were almost the same ($p>0.05$). Uncooked meatballs had a fat content ranging from 8.5 to 9.1%, cooked meatballs had a fat content ranging from 7.9 to 8.8%. Incorporation of legume flours increased protein contents of meatballs ($p<0.05$). Extended with BBF and CF slightly increased the ash content of raw meatballs. Meatballs formulated with rye bran had higher ash content than all meat control (Yılmaz, 2004).

Cooking slightly decreased moisture and increased protein content of meatballs. The protein content ranged from 18.8 to 21.1% for uncooked meatballs, from 19.3 to 23.5% for cooked meatballs. Fat and protein contents of meatballs were within the limits of Turkish Uncooked Meatball Standard (TSE, 1992). Extended with BC resulted lowest (18.8% uncooked, 19.3% cooked) protein content. Several researchers have found that protein content of comminuted meat products increased with the addition of soy proteins (Tömek et al. 1988), cowpea flour (Prinyawiwatkul et al. 1997).

Table 5 shows the changes in TBA values. On 0^{th} day, no differences were observed between the TBA values of meatball samples. In other investigation periods meatballs with BBF and CF had similar TBA values and these values were lower than the TBA values of meatballs with LF and meatballs with R. Antioxidative properties of some fruits and vegetables have been showed in various meat products (Ulu, 2004; Mansour and Khalil, 2000). At the end of the storage period all meatballs had TBA values in consumable limits and were 2.11 mg ma/kg for BBF, 1.99 mg ma/kg for CF and 2.88 mg ma/kg, 2.55 mg ma/kg for LF and R treatments respectively. Ulu (2004) concluded that 0.2% soya protein isolate was effective retarding lipid oxidation in cooked meatballs.

Unsaturated fatty acids were in similar amounts in all formulations. Our results are similar to those determined by Yılmaz (2004), Yılmaz and Dağlıoğlu (2003) for unsaturated and saturated fatty acids. Meatballs extended with BBF and CF had similar concentrations of total saturated and unsaturated saturated fatty acids. Meatballs with LF and R had similar concentrations of total saturated and unsaturated fatty acids. There were significant differences between the amounts of total polyunsaturated fatty acids of meatball samples; samples with rusk had the highest amount of total polyunsaturated fatty acids.

Conclusions

This study suggests that legume flours (blackeye bean, chickpea and lentil) can be successfully used in meatball formulations as extenders. Protein content of meatballs increased with the addition of legume flours. Legume flours are potential source of non-meat protein for meatballs. Blackeye bean flour and chickpea flour slightly retarded oxidative changes during frozen storage. There is significant difference among the meatball samples in respect to fatty acid composition.

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Table 1 Chemical composition of uncooked and cooked meatballs

Sample	Raw Meatballs				Cooked Meatballs			
	Moisture %	Fat%	Protein%	Ash%	Moisture %	Fat%	Protein%	Ash%
BBF	63.1±1.04[A]	8.7±0.28	22.0[b]±0.52	3.6±1.46	58.2±0.43	8.8±0.29	23.2[b]±0.15	2.7±1.16
CF	64.1±0.03	8.5±0.09	21.1[b]±0.34	3.2±0.02	57.4±0.35	7.9±0.71	23.5[b]±0.48	2.6±0.61
LF	65.0±0.54	9.1±0.36	21.1[b]±0.09	2.8±2.76	60.5±1.86	8.7±0.48	23.5[b]±0.43	2.8±1.10
R	63.0±0.42	8.5±0.59	18.8[a]±0.52	2.7±0.05	59.7±0.05	8.3±0.77	19.3[a]±0.26	2.8±1.12
P	NS	NS	0.012	NS	NS	NS	0.082	NS

BBF: blackeye bean flour, CF: chickpea flour, LF: lentil flour, R: rusk, [A]Standard deviation, NS: non-significant, [a-b] Different superscripts in the same column indicate significant differences ($p<0.05$)

Table 2. Fatty acid composition of meatball samples.

Fatty Acids	BBF	CF	LF	R
Saturated	47.9[a]	47.4[a]	39.6[b]	36.0[b]
Monounsaturated	23.9[c]	47.3[a]	37.9[b]	27.5[c]
Polyunsaturated	28.2[b]	5.5[c]	22.5[b]	36.5[a]
Unsaturated	52.1	52.8	60.4	64.0
Unsaturated/Saturated	1.1	1.1	1.5	1.8

BBF: blackeye bean flour, CF: chickpea flour, LF: lentil flour, R: rusk, [a-c] Different superscripts in the same column indicate significant differences ($p<0.05$)

Table 3 Changes in TBA values of meatball samples (mg ma/kg)

Sample	0th day	1st Month	2nd Month	3rd Month
BBF	0.67[x]±0.76[A]	1.02[ay]±0.95	1.67[az]±0.49	2.11[az]±0.63
CF	0.75[x]±0.12	1.13[ay]±0.22	1.55[ay]±0.63	1.99[ay]±0.91
LF	0.82[x]±0.23	1.88[by]±0.67	2.27[by]±0.07	2.88[bz]±0.21
R	0.63[x]±0.88	1.88[by]0.55	2.11[by]±0.56	2.55[by]±0.14

BBF: blackeye bean flour, CF: chickpea flour, LF: lentil flour, R: rusk, [A] Standard deviation, [(a-b)] treatments within the same storage condition with the same superscripts are not different, [(x-z)] storage conditions within the same treatment with the same superscripts are not different.

DEVELOPMENT OF BEEF BURGER MEAT PATTIES ENRICHED IN CLA

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Background

Conjugated linoleic acid (CLA) refers to a group of fatty acids comprised of different positional (placement of the C=C double bonds along the carbon chain) and geometrical (configuration around the C=C double bonds) isomers of linoleic acid (cis-9(Z), cis-12(Z) C18:2). Conjugated fatty acids are predominantly found in the fats and tissues of ruminants (C18:2 and C18:3) with a few plant (C18:3) and marine (C18:4, C20:4, C22:6) sources also known (Parodi, 1977; Chin et al., 1992; Cahoon et al., 1999; Hamberg, 1992). Typically over 80% of the CLA in dairy products is composed of a single isomer, cis-9(Z), trans-11(E) C18:2 (Sehat et al., 1998). Much interest has been generated for CLA in recent years due to the demonstration of a number of health-promoting biological activities in animals, including anticarcinogenic, antiatherogenic, antidiabetogenic, and immunomodulatory effects. In this regard CLA may be a very important functional food ingredient that food industries will need to take advantage of in future developments if the same health benefits are shown to occur in humans.

Objectives

To demonstrate how natural ingredients increase the CLA content in ruminants through dietary intervention and the use of the adipose fat depot as a functional ingredient enriched in CLA for the production of other food products such as burgers.

Materials and methods

Sixty Charolais crossbred heifers (mean initial bodyweight = 333kg, s.d. 39.90) were blocked by initial bodyweight and, within block, were randomly assigned to one of four dietary treatments (n = 15): 1) indoor, silage/concentrates (control, SC); 2) unsupplemented grazing (G); 3) restricted grazing plus 2kg/head/day of linseed oil-enriched meal (LSG) and 4) restricted grazing plus 2 kg/head/day of sunflower oil-enriched meal (SFG). Concentrates and grass allowances were monitored at three-week intervals during a 5-month experimental period to achieve similar carcass weights across the treatments. Animals were slaughtered at a commercial facility, carcasses were chilled for 48 h at 4°C, and the *M. semimembranosus* (SM) and subcutaneous adipose tissue (SC) were excised from each carcass.

The fatty acids (FAs) were extracted and saponified from tissue samples in 6mL 5M KOH in methanol/water (50:50) at 60ºC for 1 hour and methylated using trimethylsilyl-diazomethane in methanol:toluene (2:1 %v/v) at 40ºC for 10 min based on a modified method by Elmore *et al*. (1999). Separation of fatty acid methyl esters (FAMEs) was performed on a Varian CX3400 GC, using a BPX-70 column (120m x 0.25mm i.d., 0.2□m film thickness, SGE, Australia) with a programmed temperature ramp. Injector and detector were set at 270°C and 300°C respectively. The carrier gas was hydrogen set to a flow rate of 1.6ml/min, measured at the initial temperature and using a split ratio of 50:1. FAMEs were identified according to similar peak retention times using standards (Sigma Chemical Co. Ltd., Poole, U.K.), and quantified according to the use of an internal standard ($C_{23:0}$ methyl ester) with its addition prior to saponification. The data was analysed as a randomized block design using MiniTab 14.

Adipose tissue was evaluated for its content of beneficial fatty acids, and, its suitability as an enriched source of CLA that could be used in product development. Beef burgers were produced from the *M. semimembranosus* and the CLA-enriched fat. Fatty acid analysis and sensory evaluation was carried out on these burgers to determine the effect if any on flavour and textural quality attributes

Results and discussion

Fatty Acid Results:

A processed meat product was produced and made available for CLA and fatty acid determinations. The *M. semimembranosous* was used in conjunction with carcass fat to produce beef burgers. Burgers were compared with respect to the initial dietary treatments (table 1). Of interest is the following: 1. the fatty acid content of burgers is quite different to the lean muscle (because of the high percentage of adipose tissue present in the burgers) – long chain polyunsaturates are less abundant (C20:4, C20:5, C22:5, C22:6) but a relatively high amount of C18:1 trans-11 (TVA) and CLA is present; 2. quite marked treatment affects are observed illustrating the production of burgers high in certain fatty acids such as CLA (table 1), diets rich in linoleic acid such as the sunflower oil supplemented diet drive the synthesis of CLA cis-9, trans-11 and C18:1 trans-11 (trans vacenic acid). The results also illustrate a large difference compared to conventional indoor silage-based diets (the content of CLA in silage - control Vs sunflower oil supplemented diet differs by more than 400%!, figure1).

Of the four different dietary treatments the linseed diet contained the most polyunsaturates (PUFA), followed by the sunflower diet and pasture diet with the silage diet containing the least amount of PUFA. For the total saturated fatty acids (SFA) the positions were reversed with the control diet containing the most SFA, followed by the pasture diet and sunflower diet while the linseed diet contained the least amount of SFA. The P:S ratio was highest for the linseed diet and lowest for the silage diet. The n-6: n-3 ratio was lowest for the pasture based diet, followed by the linseed, then silage and finally sunflower oil diets.

Sensory Evaluation:

An inhouse, trained panel of 20 people from different backgrounds was established to access the following sensory attributes: texture (hardness, softness), juiciness, and flavour quality. In addition they were also asked to give an overall acceptability score for the various samples. Samples were graded on a categorical scale from 1 to 6, were 1 is generally least favoured, to 6 the most preferred. In general, higher preference scores were associated with the pasture and oil-supplemented diets. Significant differences ($P< 0.05$) were observed between the silage and the pasture and oil-supplemented diets. It must be stressed however, that these results do not imply that meat from the silage based diet tasted bad or was unacceptable, merely that the panellists showed a trend favouring burgers from the other dietary treatments.

Conclusions

Supplementing grazing animals with plant oil-enriched concentrates resulted in a further beneficial effect on the fatty acid composition of muscle and fat compared to grazing alone. Sunflower oil was more effective in increasing the concentration of CLA and TVA, but had a negative effect on the n-6:n-3 ratio, while linseed oil supplementation had a less pronounced effect on the CLA concentration than sunflower oil.

Beef burgers enriched in CLA were successfully produced and sensory evaluation indicated that there was no detrimental effect on burger flavour and other sensory qualities. In fact, many sensory attributes are improved by pasture and oil supplementation (based on burger sensory analysis).

The results clearly show the suitability of fatty tissue from oil-supplemented diets to be a particularly rich source of CLA. An interesting feature here is the difference in the qualitative fatty acid profile and the relative abundance of individual fatty acids when comparing adipose tissue and lean muscle tissue. The differences presumably reflect the two distinct biological roles these tissues play with respect to their lipid profiles. Adipose tissue is a predominantly storage depot of fat, while muscle adjusts its content of fatty acids (predominantly the phospholipids fraction) to suit the functioning of its membrane systems (calcium ion storage and electrical impulse conduction).

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	Fatty acids (mg 100g^{-1} Burger)				
	SC	G	SFG	LSG	P
Palmitic acid (C16:0)	8417 (613)	6883 (965)	6334 (707)	6427 (810)	***
Stearic Acid (C18:0)	3440 (222)	3596 (594)	3685 (401)	3488 (433)	NS
TVA (C18:1*trans*11)	380 (40)	1051 (165)	2510 (263)	1942 (250)	***
Oleic Acid (C18:1)	9357 (683)	8341 (1126)	8050 (778)	7482 (911)	NS
Linoleic Acid (C18:2)	346 (26)	294 (41)	403 (36)	326 (37)	***
Linolenic Acid (C18:3)	138 (10)	227 (31)	148 (15)	193 (27)	***
CLA cis9,trans11	153 (13)	328 (47)	647 (59)	454 (61)	
PUFA	801	1208	1498	1686	***
SFA	13010	11614	11079	10981	NS
P:S Ratio	0.06	0.10	0.14	0.15	***
n-6:n-3 Ratio	2.30	1.18	2.37	1.56	***

Table 1: Results of selected fatty acids from burgers produced from animals fed different diets. SC = silage/concentrates, G = pasture only, SFG = sunflower oil supplemented, and LSG = linseed oil supplemented. Values are expressed as mean +/- standard deviation.

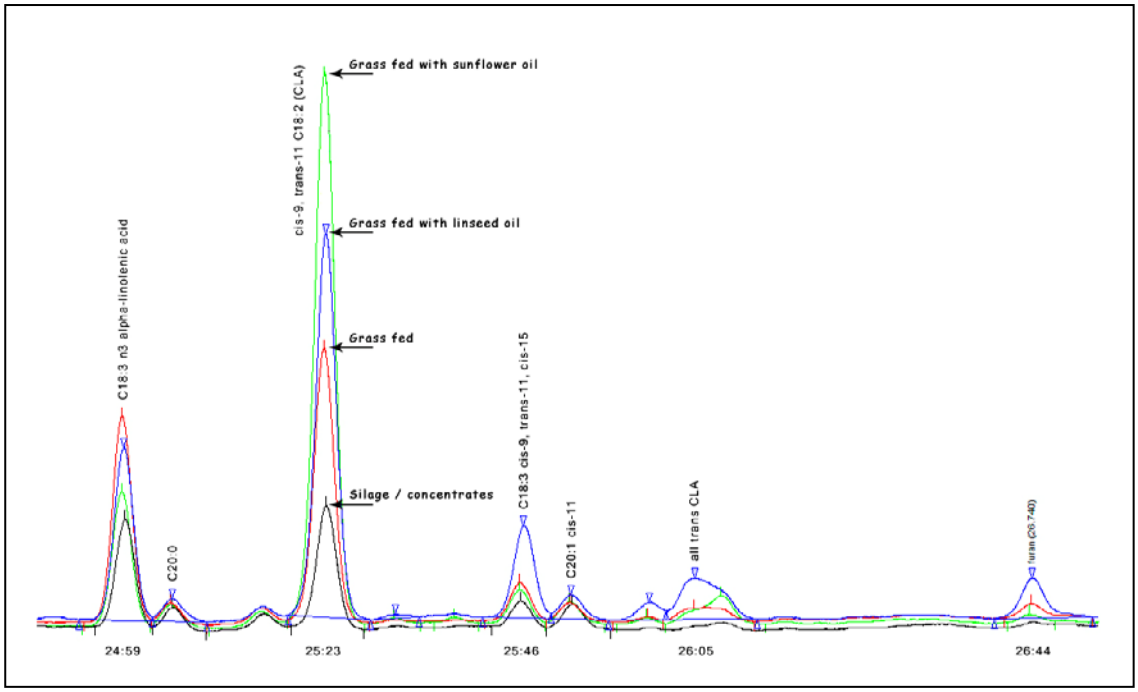

Figure1: Gas chromatogram illustrating the relative differences in the CLA peak (cis-9, trans-11 isomer) from burgers produced from animals on different diets.

EFFECTS OF *POLYGONUM HYDROPIPER* L. EXTRACTS ON THE QUALITY OF CHILLED BEEF STEAK

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Abstract

This study was undertaken to examine the effects of *Polygonum hydropiper* L. extracts on the quality of Australian and Indian beef sirloin steaks. The use of 2000ppm extracts were observed to reduce lipid oxidation during 18 days chilled storage while pH values decreased during storage time in both types of meat, treated (K1 & K2) and untreated (S1 & S2). The application of *Polygonum hydropiper* L. extracts also resulted in the lower hardness values. No significant differences were observed in the L (lightness) value , however there was a decrease in a (redness) value for all samples . As for b (yellowness) value not much difference were detected except for Australian untreated beefsteak which increased drastically after 18 days of chilled storage.

Keywords: *Polygonum hydropiper;* extracts; beef steak; chilled

Introduction

Buffalo meat (Indian beef) are being imported in large volume by Malaysia while higher quality beef cuts are imported in smaller volume from Australia. Steaks from sirloin, striploin and ribeye muscles are generally tender and are quality cuts preferred by the consumers. However restaurants and hotels serving steaks are reluctant to use Indian fillets, referring to poor quality such as toughness, chewiness, low sensory acceptance and poor microbial quality related to meat imported from India. It is with this unconfirmed factors that this study research into the effects of plant extracts to improve certain quality attributes related to the consumers perception of Indian beef. Australian beef fillets are well received by local consumers as well as foreigners who associate beef from Australia as superior and of high quality.

Materials and methods

Preparation of beef steaks

Frozen (-18^{o}C) *longisimmus dorsi* muscles in the form of striploin or sirloin were purchased from a local importer (Lucky Frozen Sdn. Bhd.) in Kuala Lumpur, Malaysia. Beef were cut into pieces and marinated with salt (1.5%) and pepper (2%) for control, namely K1 (untreated Indian beefsteak) and K2 (untreated Australian beefsteak). 2000ppm of *Polygonum hydropiper* L. extracts were applied topically to steaks by dry rubbing the external surface of the steaks on both side for sample S1 (Indian beefsteak) and S2 (Australian beefsteak). The samples were stored at 5 $\pm$ 1^{o}C for 18 days.

pH determination

Duplicate beef samples were periodically removed from storage and homogenized in distilled deionized water (1:10 dilution). Homogenates were filtered through a Whatman No. 1 filter paper to obtain clear filtrate for pH measurement.

Colour measurement

Surface colour of sirloin steaks was determined for L (lightness), a (redness) and b (yellowness) using a Minolta Chromameter CR-100.

Texture Analysis

Textural characteristics of sirloin steaks were analyzed using a Warner-Bratzler shear machine.

Lipid Oxidation Analysis

Thiobarbituric acid numbers (TBA) was determined following the distillation method described by Tarladgis et al (1960) with little modification by Rhee (1978).

Statistical Analysis

The data were analyzed using the Statistical Analysis Systems (SAS) program version 6.12 (SAS 1995). Treatments showing significant differences ($p<0.05$) were subjected to the Duncan's Multiple Range Test.

Results and discussion

Fig 1 shows the pH values for the beef steaks stored at chilled temperature for 18 days. There was a significant decrease in pH values for all samples after chilled storage for 18 days. However there were no significant difference in pH values between the treated and untreated (control) samples for both types of meat. The lower pH values for all samples during the chilled storage could be due to production of free fatty acids from the phospholipids fraction and the separation of free fatty acid from the triacylglycerol (Rhee et al. 1977). The pH of fresh meat can be influenced by the presence of bacteria and may reflect the relative differentiation between the presence of gram positive or gram negative bacteria. Organic acids, produced by gram positive bacteria, decreased the pH of meats, whereas amines produced by gram negative bacteria increase pH (Lefebvre et al., 1994).

Colour changes in beef steaks during chilled storage were shown in Figures 2, 3 and 4.
There were not much changes in the lightness of the samples except for untreated Australian beef steak which increase after 18 days of chilled storage. Values for redness were found to decrease significantly after 18 days of storage in all samples. A loss of surface redness in all samples can be attributed to decrease in dissolved oxygen, due in part to the utilization of oxygen by psychrotrophic bacteria, and the participation in oxidative reactions such as lipid oxidation (Peter & David, 2002). As for yellowness values no significant difference were found in all samples except for K2 yellowness which increased drastically during storage.

Figure 5 showed the results for texture (gF) for the four samples. Warner-Bratzler shear measurement showed an increase in values indicating increase in toughness of all samples. This could be attributed to the moisture loss during the chilled storage.There was significant difference in toughness between the control K1 and K2 whereby K1 was tougher than K2. For samples treated with *Polygonum hydropiper* L. extracts (S1 & S2) the toughness was significantly lower than the control (K1 & K2). Lipid soluble antioxidant from plant extracts can maintain integrity of muscle fibres and reduce moisture loss (Mitsumoto et al., 1995).
Rancidity of meat products are indicated by an increase in the content of malonaldehyde normally measured by TBA method. In this study there was a significant increase in TBA values in all samples as a result of the 18 days chilled storage (Fig. 6). However the values of treated samples (S1 & S2) were significantly lower than the controls (K1 & K2) though K1 showed higher values than K2. Taylor (1987) suggested prooxidants such as iron and copper from water and spices to contribute to increase in rancidity of meat products. Besides, the process of cutting and mixing during preparation of samples also increased oxidation in meat. Lower TBA values in treated samples for both types of meat indicated that *Polygonum hydropiper* L. extract was effective in inhibiting the formation of malonaldehyde, thus delaying lipid oxidation of the chilled beef steaks. The extracts was known to contain ten flavonoid compounds that possess strong antioxidative activity (Zhao et al., 2003).

Conclusions

The addition of *Polygonum hydropiper* L. extracts to the Australian and Indian beef steaks was found to improve the quality attributes such as texture and oxidative changes. The toughness was reduced when compared to the untreated samples for both types of meat. The oxidative process increased at a slower rate when compared to the untreated samples after 18 days of chilled storage at 5^{o}C.

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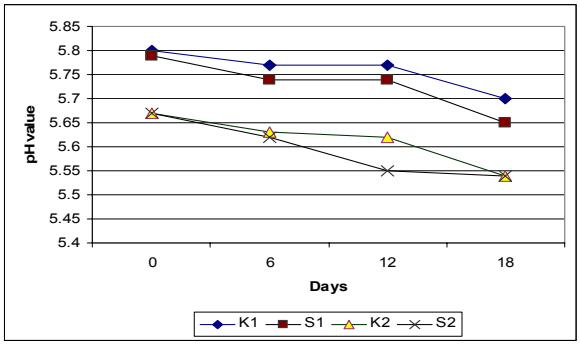

Fig. 1: pH values of chilled storage beef steaks

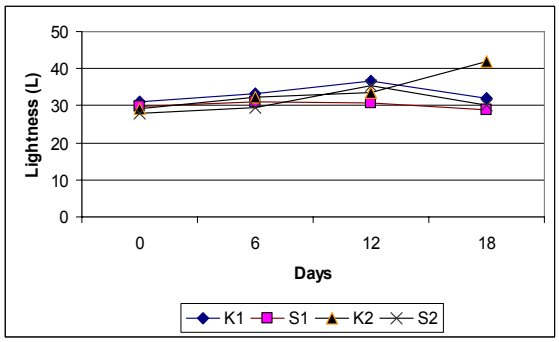

Fig. 2: Lightness (L) values of chilled storage beef steaks

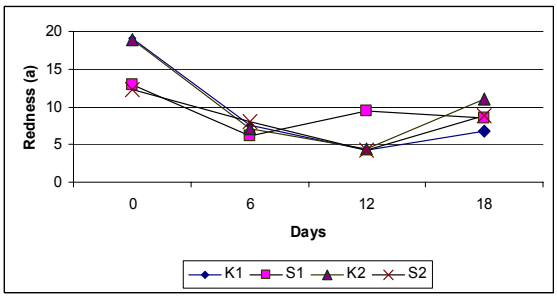

Fig. 3: Redness (a) values of chilled storage beef steaks

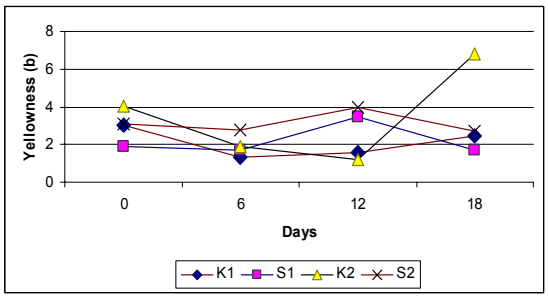

Fig. 4: Yellowness (b) values of chilled storage beef steaks

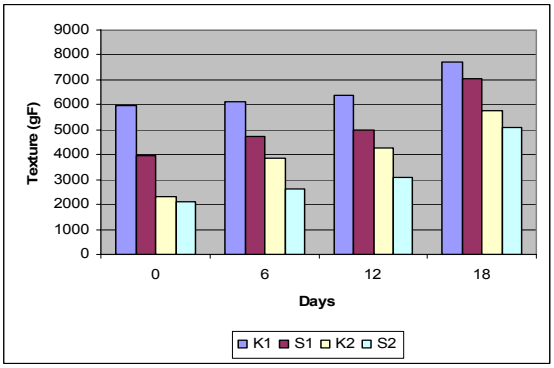

Fig. 5: Texture values for chilled storage beef steaks

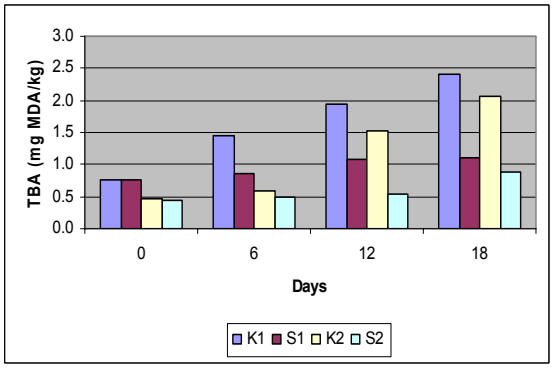

Fig. 6: TBA values for chilled storage beef steaks