

## BENCHMARKING VALUE IN THE PORK SUPPLY CHAIN: PROCESSING CHARACTERISTICS AND CONSUMER EVALUATIONS OF PORK BELLIES OF DIFFERENT THICKNESSES WHEN MANUFACTURED INTO BACON

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## Background

Bacon consumption and use has seen extensive growth throughout the 1990's. This trend has had an important effect on the pork industry by increasing demand and value of fresh pork bellies. The 1992 Pork Chain Quality Audit (Cannon *et al.*, 1992) reported that 10% of bellies were too thin for bacon production and an additional 2% were too soft/oily to be used in bacon manufacturing. Quality defects identified by packers, such as belly thickness, often manifest substantial losses in profitability at the processor level due to reduced processing yields and a greater percentage of inferior products when sliced.

Improvements in swine genetics, nutrition, and management decisions have produced pork carcasses with 19% heavier hams and 21% heavier loins than carcasses from 10 years ago, whereas belly weights have remained constant (Stetzer and McKeith, 2003). Mandigo (2002) reported that bellies from current market swine contain approximately 29% less fat compared to bellies from swine 40 years ago. These changes have resulted in decreases in the thickness of bellies. This is a cause of concern because thicker bellies have been found to have higher processing yields than thinner bellies (Jabaay *et al.*, 1976). Nonetheless, West *et al.* (1973) and Jabaay *et al.* (1976) have shown that consumers prefer leaner bacon, which is often derived from thinner bellies that contain less fat.

## Objectives

To understand the relationship between belly thickness, processing yields and consumer preferences so that bacon processors may develop raw material specifications that maintain processing efficiency without compromising customer satisfaction.

#### Materials and methods

Fresh pork bellies (IMPS #408; NAMP, 1997; USDA, 1997) were selected from 500 kg lots consisting of bellies ranging in weight from 5.4 to 7.3 kg at a commercial bacon manufacturing company. Bellies (n = 96 per group) were sorted subjectively into "thin" (approximately 2.0 cm), "average" (approximately 2.5 cm), and "thick" (approximately 3.0 cm) belly thickness classifications by three evaluators. Sorting procedures were validated by measuring the thickness of all bellies using digital calipers at three locations (blade end, center, and ham end) along the dorsal and ventral edges. Bellies were skinned, and skin weights were recorded to calculate skinning yield. After skinning, bellies were injected to 110% of raw weight with a curing solution specific for that commercial facility and chilled at 2°C for 24 hrs. Following storage, bellies were smoked, chilled, and pressed into uniform rectangles according to standard industry procedures. Weights were recorded during all phases of production and used to calculate processing yields.

Bellies were sliced into uniform strips (approximately 3 mm and weighing 28 g) using standard industry equipment. During slicing, bacon slices were classified according to their characteristics for secondary lean and slice thickness. The most valuable slices, "#1 slices," were those that met requirements for secondary lean characteristics (*M. cutaneous trunci* greater than 50% of the width of the slice) and appropriate slice profile thickness (no measurement less than 1.9 cm in profile thickness at any point). The less valuable slices, "#2 slices," were those that had insufficient secondary lean characteristics (*M. cutaneous trunci* less than 50% of the width of the slice) or inappropriate slice thickness (a measurement less than 1.9 cm in profile thickness at any point). Those slices not meeting "#1 slice" or "#2 slice" characteristics and slices

from the cranial or caudal ends of bellies were classified as "ends and pieces." Weights of each component were recorded and slicing yields were calculated. During slicing, approximately 500 g of bacon slices were collected from the center portion of every third belly (n = 32 per group). Bacon slices were vacuum-packaged and shipped to the University of Illinois for further testing.

Consumer evaluations of bacon were conducted at the University of Illinois. Bacon slices were placed on racks and cooked in convection ovens set at 232°C for 12 min. To ensure even cooking throughout slices, racks were rotated after 6 min. Bacon slices were removed from racks, blotted with paper towels to remove excess grease, and placed on paper plates as individual slices. Participants were recruited from residents of the Champaign-Urbana, Illinois area and asked to complete a basic demographic worksheet. Consumers (n =120) were placed in individual sensory evaluation booths and asked to evaluate bacon slices, served in a random order, for flavor, fattiness, saltiness, crispiness, leanness, and pinkness. In addition, consumers were asked to evaluate the visual appearance of uncooked bacon slices. Bacon slices were laid out individually under cool white fluorescent light against a white background and placed in separate evaluation cubicles to prevent visual comparison between slices. Slices were selected randomly from packages representing each thickness group, and slices were replaced periodically throughout the evaluation timeframe. Also, purchase intent was recorded for both taste and appearance. Sensory and visual evaluations were scored on 5-point scales. For flavor, fattiness, saltiness, crispiness, leanness, and pinkness, scales were 1 = much too little, 2 = somewhat too little, 3 = just right; 4 = somewhat too much, 5 = much too much. For taste and visual appearance, scales were 1 = extremely unacceptable, 2 = moderately unacceptable, 3 = neither, 4 = 1moderately acceptable, 5 = extremely acceptable. For purchase intent for taste and appearance, scales were 1 = definitely, 2 = probably, 3 = might, 4 = probably not, 5 = definitely not.

Proximate composition was determined using raw, and cooked (n = 5 slices per group) bacon slices. Samples were ground and moisture content determined using AOAC (1995) approved methodology, and fat content of samples was determined by a chloroform-methanol method.

Data were analyzed using SAS (SAS Institute, Cary, NC). Descriptive statistics and frequency distributions were generated using the PROC Means and PROC Freq procedures, respectively. Frequency distributions for consumer sensory responses were tested for significance (P < 0.05) using chi-square analysis. Analysis of variance was performed using the PROC GLM procedure with belly thickness group tested as the main effect. When main effects were determined to be significant (P < 0.05), least squares means were generated and separated using a pairwise t-test (pdiff option).

## **Results and discussion**

Objective measures of belly thickness were recorded to validate subjective selection procedures. Mean thickness between groups were different with approximately 4 mm separating "thin" from "average" bellies, and 5 mm separating "average" from "thick" bellies. "Thin" bellies had slightly higher skinning losses than "average" or "thick" bellies, but this is likely a function of green weight as the weight of skins collected from each thickness group was nearly identical. Likewise, bellies from the "thin" thickness group had the highest cooking shrink, which also was probably related to lighter belly weights because all bellies were subjected to the same smoking and cooking cycle. Brewer *et al.* (1995) reported a strong correlation (r = 0.70) between belly thickness and raw belly weight. Currently, most bacon processors purchase bellies by weight This possibly accounts for a portion of the variation in belly thickness, but there still is categories. substantial variation within weight ranges that may be best controlled by sorting bellies based upon thickness measures in lieu of or in concert with weight. By sorting bellies on thickness, processors may be able to regulate cooking shrink if all bellies in a cook cycle are of a similar thickness. Within the weight ranges included in this study (approximately 5 to 7 kg), which accounts for approximately 42% of available bellies (Stetzer and McKeith, 2003), we were able to find the variation in raw materials. "Thick" bellies had the highest final yield, which was approximately 2.3 percentage points higher than the yields for bellies from the "thin" and "average" groups. No differences in final yields were observed between bellies from the "thin" and "average" groups.

Bellies from the "thin" group had the lowest slicing yields as evidenced by the lowest percentage of #1 slices (i.e., more valuable product), and the highest percentage of #2 slices (i.e., less valuable product), and "ends

and pieces." Thickness groups had an equal percentage of slices classified as #2 product because of inadequate secondary lean (7.2% for "thin," 7.5% for "average," and 7.1% for "thick"). This is not surprising because we would expect the *M. cutaneous trunci* to remain relatively proportional to the width of the belly. Thus, "thin" bellies had a higher yield of #2 slices because those slices were too thin in profile to be classified as #1 slices (1.5% for "thin," 0.3% for "average," and 0.2% for "thick").

Demographic information provided by consumers showed that 80% of participants were 24 years old or older, 43% had an income of \$35,000 or higher, and 60% were female. Consumer evaluations of bacon flavor showed that consumers gave similar responses for "thin" and "average" thickness bellies, whereas respondents indicated that slices of bacon from "thick" bellies lacked bacon flavor. Nearly 60% of consumers indicated that slices of bacon from "thick" bellies was appropriate, whereas 38% and 43% of consumers indicated that slices of bacon from "thin" and "average" thickness bellies were too salty. Belly thickness may have an effect on flavor attributes with thicker bellies diluting flavors more than "average" or "thin" bellies. In contrast, Brewer *et al.* (1995) found no differences in bacon flavor or saltiness as belly thickness increased. A consumer panel was used in this study, whereas a trained sensory panel was used by Brewer *et al.* (1995). This may explain why a discrepancy in findings exists. Generally, consumers gave similar responses for fattiness evaluations for each thickness group, which may be expected because bacon is approximately 50% fat.

"Thin" bellies had a greater frequency of responses indicating less crispiness compared to bacon slices from "average" and "thick" bellies. It appears bacon from "thin" bellies may require greater cooking times to achieve the crispiness level consumers desire. Compositional data showed that bacon slices from "thin" bellies contained approximately 7% higher moisture than bacon slices from "average" or "thick" bellies, which may have provided extended evaporative cooling during cooking, and prevented slices from becoming as crispy as slices from "average " and "thick" bellies. This could have ramifications on precooked bacon slices because processors may need to adjust cooking parameters for slices from "thin" bellies to meet the crispiness expectations of consumers.

Generally, responses for overall taste were similar for all belly thickness groups. "Thin" and "thick" bellies had a slightly greater frequency of responses indicating unacceptable taste than slices from "average" bellies. This may be related to fatty acid composition of "thin" bellies, which are usually softer and/or oilier, may have a higher percentage of unsaturated fatty acids, whereas "thick" bellies, which are usually firmer, may contain a greater percentage of saturated fatty acids. In agreement with overall taste data, consumers showed slightly increased likelihood of purchasing bacon from "average" thickness bellies based on taste compared to "thin" or "thick" bellies, with 65.6% of consumers indicating they would "definitely" or "probably" purchase bacon from "average" thickness bellies compared to 50.5% and 48.3% for bacon slices from "thin" or "thick" bellies, respectively. Likewise, greater than 25% of consumers indicated they would be unlikely (i.e., "probably not" or "definitely not") to purchase bacon manufactured from "thin" or "thick" bellies, respectively, because of taste compared to 17.3% for "average" bellies.

Most consumers (73.7%) gave responses indicating that the lean to fat ratio for bacon slices from "thick" bellies was too low (i.e., too much fat in relationship to the amount of lean), whereas greater than 50% of participants indicated that the lean to fat ratio in bacon slices from "thin" and "average" bellies was "just right". Similarly, Brewer *et al.* (1995) observed that as belly thickness increased, sensory assessments of lean-to-fat ratio decreased.

Approximately 70% of consumers indicated that the pinkness of lean in bacon slices from "thin" and "average" bellies was "just right," whereas 56.6% of consumers indicated that the pinkness of bacon slices from "thick" bellies was less than ideal (i.e., "much too little" and "somewhat too little"). It is unclear if the fatness of bacon slices from "thick" bellies "diluted" the pinkness of color in those slices or if it is more difficult to distribute a curing solution in those bellies, resulting in less pinkness.

Consumer responses for visual acceptability indicated that raw bacon slices from "average" (72.4%) and "thin" (78.7%) bellies were acceptable (i.e., "moderately acceptable" or "extremely acceptable"). In contrast, a majority of consumers (56.3%) found slices of bacon from "thick" bellies visually unacceptable. Purchase intent based on visual appearance closely mimicked consumer responses for visual appearance.



For bacon slices from "thin" and "average" bellies, 62.3% and 59.4%, respectively, of consumers indicated that they were likely to purchase those products. In contrast, 62.4% of consumers indicated that they were unlikely to purchase bacon manufactured from "thick" bellies. Our findings agree with the results of Jabaay *et al.* (1976) and West *et al.* (1973) who reported that consumers preferred the appearance of leaner bacon.

Compositional differences were observed in bacon slices from different thickness groups. Raw bacon manufactured from "thin" bellies had the highest moisture content (47.9%) and less fat (36.2%) than raw bacon from "average" (40.5% moisture; 46.4% fat) and "thick" (40.4% moisture; 46.3% fat) bellies. Brewer et al. (1995) reported that as bellies became thicker, moisture content decreased and fat content increased. Compositional differences between thickness groups were similar for cooked bacon. Cooked bacon from "thin" bellies contained less fat (34.7%) than cooked bacon from "average" (42.5%) and "thick" (43.5%) bellies. It is surprising that the composition of bacon from "average" and "thick" bellies was so similar because consumers identified bacon from "thick" bellies as having the lowest lean-to-fat ratio and discriminated against it because of its excessive fatness.

## Conclusions

Belly thickness influenced the processing yields and consumer reactions to the acceptability of bacon. "Thick" bellies offer processors the greatest processing yields, however, it is likely that consumers will discriminate against the resulting products because of excessive fatness. Consumers viewed bacon from "average" and "thin" bellies very favorably, however, working with "thin" bellies may reduce industry profitability because of lower processing yields. Swine producers and geneticists should focus on producing swine with bellies of "average" thickness.

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## EFFECT OF VACUUM PACKAGE STORAGE ON PORK PURGE AND COLOR

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#### Background

Vacuum packaging is used to extend the shelf life of fresh pork and is known to preserve the microbiological integrity of the product for up to 12 weeks (Jeremiah et al, 1995). Purge is a consequence of chilled storage and is seen as a defect when the Japanese customer opens the vacuum package. Factors such as pre-slaughter management, stunning procedures, chilling rates and holding temperatures will affect drip and color. Genetics play an important role in this regard and this data set will investigate the effect that breed has on the meat quality of vacuum packaged loins.

#### **Objectives**

The objective of this study was to investigate the effect of background genetics on meat quality of vacuum packaged loins as assessed by measuring purge and color of the meat.

#### Materials and methods

Pig breeds used in this trial originated from the PIC genetic nucleus in Kipling Saskatchewan, Canada and included the following basic genotypes: Landrace (Land), Large White (LW), Duroc, White Duroc (Wdur), Berkshire (Berk), Hampshire (Hamp), Synthetic line (Syn), Berkshire x Hampshire Cross (BH), hal-negative Pietrain (Piet<sup>-</sup>), hal-positive Pietrain (Piet<sup>+</sup>). A section of the longissimus muscle from the right side between the 5<sup>th</sup> and 10<sup>th</sup> ribs was cut into four equal size portions and vacuum packaged. Packages were then randomly allocated to one of four storage intervals (0, 4, 6 and 8 weeks) and stored at  $2^{\circ}C (\pm 0.5^{\circ}C)$  for the designated storage interval. Following storage, the packages were weighed and opened. The meat samples were removed, blotted dry and weighed. The packages were air dried and weighed. Purge was calculated by difference. Samples were then placed on Styrofoam trays, over-wrapped with an oxygen permeable film (Vitafilm Choice Wrap), and allowed to bloom for 1 hour prior to being evaluated by a 5-member trained and experienced sensory panel for muscle color (5 point descriptive scale), surface discoloration (7 point descriptive scale), and retail appearance (7 point hedonic scale). The packages were then opened and the samples were evaluated for off-odors (5 point descriptive scale) and odor desirability (5 point hedonic scale) (Jeremiah and Gibson 1995). CIE L\*, a\* and b\* were measured in triplicate on each muscle section at each storage interval using a Minolta CR-300 reflectance meter (illuminant - C, observer angle - 2°). Statistical analysis was performed using the GLM procedure of SAS (2001).

#### **Results and discussion**

Percent purge loss measurements by genotype over storage time are presented in Figure 1. In general, there is known to be a positive relationship between percent purge and the length of storage (Apple et al. 2001; Apple et al. 2002; Kim et al. 1998,). Statistically there is a significant interaction between genotype and week in storage. Overall, the Berk shows the lowest level of purge while the Piet<sup>+</sup> has the highest percent purge values. The high percent of purge of the Piet<sup>+</sup> was expected due the presence of the halothane gene (Apple et al., 2002). The low purge level of the Berk is expected since it usually displays low rates of muscle lactic acid production that would result in a minimum amount of protein denaturation and better WHC (NPPC, 1995). In terms of time dependent effects on purge, the Dur, Syn and BH are the only lines that show a significant increase between week 4 and week 6. All the other genotypes show no significant increase in purge after after week 4. This observation indicates that accumulated purge has reached a maximum at week 4 and that in order to measure rates of accumulation in packages, purge may well be better evaluated within the first 4 weeks of packaging.

L\* measurements are presented in Figure 2. In general, as the storage time increases the meat tends to have a paler appearance. This trend is generally and clearly displayed in the results where, at packaging time (Time



0), L values for all genotypes were below 50, and then rose sharply to above 51 by Week 4 (P<0.05). The magnitude of increase after 4 weeks in storage was then minor for all genotypes. Overall, Piet<sup>+</sup> was the palest and Syn along with Land and LW were on the other end of the spectrum, showing the darkest loins. In general, these evaluations confirm the trend towards lighter meat color for all genotypes (Apple et al. 2001; Apple et al. 2002). Along with this increase in L\* (paleness), both a\* and b\* increased with storage time for all genotypes (results not shown). The increases in a\* and b\* translate to increases in chroma and hue angle, which implies an increase in color intensity and a shift toward the yellow but within the red part of the spectrum.

The changes in purge and color during vacuum packaged storage for up to 8 weeks were accompanied by changes in a number of sensory traits (results not shown). Discoloration increased by up to 10% and retail acceptability decrease from "desirable" to "neither desirable nor undesirable". Piet+ and piet- lines were poorer at all storage times and approached "undesirable" at 8 weeks. By 8 weeks storage all genotypes had a moderate off odor which resulted in a decrease in odor acceptability (from "acceptable" to "slightly unacceptable").

A detailed characterization of the quality (pH, drip loss, etc) of the longissimus muscles of these genotypic lines can be found in the paper of Pommier et al. (2004).

## Conclusions

Genotype had a large effect on both the amount of purge in the vacuum package and on the meat color. The effects of length of storage in vacuum packages on percent pork purge loss and color are consistent with basic literature (Kim et al, 1998). In addition, the results indicate that majority of purge in the package accumulates during the first 4 weeks post packaging. Increasing storage time increased the amount of muscle discoloration, paleness, chroma, hue angle and prevalence of off odors, and after 8 weeks of storage, muscle samples decreased in retail desirability and odor acceptability as observed previously (Jeremiah et al, 1992). Any attempt to determine differences between genotypes in the rate of fluid accumulation should focus on the initial 4-week period because in most cases maximum purge is attained at that time.

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Figure 1: Effect of genotype and storage time on purge of vacuum packaged loins. (Columns within genotype with different letters differ significantly (P < 0.05)).



Figure 2: Effect of genotype and storage time on L\* of vacuum packaged loins. (Columns within genotype with different letters differ significantly (P < 0.05)).



# REARING OF PARTLY OUTDOOR PIGS IN ESTONIA AND THE QUALITY OF THE PORK

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#### Background

Meat quality depends on many factors – rearing, feeding, pre-slaughter handling, transportation, slaughtering, stress etc. Meat quality defects such as PSE-meat can be caused by poor transport and unsuitable pre-slaughter conditions.

This paper discusses studies made to investigate two different feeding and rearing systems and the pork quality in different stress conditions (low stress and high stress). Research is completed as part of the 5th Framework project "Susporkqual" and financed through EC contract No QLK 5-CT-2000-00162. Countries involved in the research were: Denmark, Poland, France, Sweden, Ireland, England and Estonia.

#### Objectives

The aim of the project for Partner 9 and 9A (Estonia) was to study: a) the interactive effects of genotype and rearing methods on stress reactivity at slaughter and on meat quality choosing country specific breeds and rearing methods, b) the effect of rearing methods on reactivity to transport; c) to find out if it is possible to produce pork more effectively by using partly out-door rearing and different feeds.

#### Materials and methods

The main difference between pigsties was, that the conventional pigsty had only an inside area  $(15,22 \text{ m}^2)$ , whilst the partly outdoor pigsty had two areas: an inside area and an outside area. Compared to conventional pigsty  $(0,7m^2 \text{ per animal})$  stocking density was higher in partly outdoor pigsty  $(1,1m^2 \text{ per animal})$ .

Piglets were from sows of Estonian Landrace breed and boars from crossbreed Landrace and Large White. All piglets were female. We selected stress-negative sows and boars on the bases of a DNA – test. Two different compositions of feed were used for indoor and outdoor pigs: a) import with soya (diet a); b) domestic (diet b) with pea + rape corn. Diets a1, b2 were given to pigs with a live weight of 30 to 60 kg and diets a3, b4 to pigs with a live weight of 60 to slaughtering. The diets were based on the general principles of pigs feed content in Estonia /5/.

Live weights (  $\sim$  30, 60 kg and pre - slaughter) of pigs, feed conversation rate (FCR) and weight gain were determined during rearing period.

Slaughtering of 80 pigs was proceeded in two different conditions: half of the pigs (n = 40) were slaughtered in Linnamäe Peekon (LP) at low transportation stress conditions (distance from farm to slaughterhouse ~ 500 m) and the other half (n = 40) in Saaremaa Meat Plant (SMP) after having left behind a distance of 200 km by lorry and ferry – high transportation stress conditions. So we had eight different groups in slaughtering: e.g. pigs from pen 1 (n = 20) were divided into two: 10 pigs were slaughtered in LP and another 10 – in SMP (Scheme).

## Methods

I. Carcass weight. Two measurements of carcass weight were made: 1.warm carcass weight; 2. cold carcass weight:

II. Back fat measurements. Linear measurements of the back fat thickness were made the day after slaughter (24 hours) at four locations.

- □ One measurement was made on the shoulder area (at the cut between the third and forth vertebra in sternum) over the middle of M. trapeziu.s (backfat 1).
- □ One measurement was made on the loin area (at the cut behind the last rib) over the middle of M. Longissimus dorsi(LD).(backfat 2)



□ Two measurements were made on the ham cut: one measuring over the middle of M. gluteus medius (backfat 3) and one measuring over the two fat layers at the thickest point at the cartilage of the hip bone. (backfat 4).

III. pH. All the pH-measurements were made in LD at the last rib with a combined electrode. The electrode was calibrated at 35 °C for the measurements in hot carcass (pH45 min) and at low temperatures for measurements in cold carcass (pH24h with ARGUS X /Sentron pH – meter.

IV. Temperature. All the temperature measurements were made with ARGUS X/Sentron pH – meter. In LD at the last rib: measurement 45 minutes after sticking – temp 45 min and 20 – 24 hours after slaughtering temp 24h.

IV. Chemical composition of meat. Chemical composition of meat: protein, fat, ash and moisture content were determinated in the 4 cm sample from LD at the last rib towards the ham. Intercalibrated methods were used. Analyses was made in the Tartu Department of the Estonian Food and Veterinary Laboratory.

V. Lean meat content in SMP was determined with Ultra – FOM 100 and in Linnamäe Peekon by the ZP – method.

## **Results and discussion**

Compared to conventional rearing partly outdoor rearing resulted in 19,45% bigger average feed intake; 9,9 % higher FCR; faster growth, described by the growth rate, higher dressing % and faster alacrity of pre slaughter live weight. All back fat measurements (except nr 4, conventional) were higher in partly outdoor reared pigs (Table 1).

The present work involves studies made to investigate how pork quality is conditioned by different stress conditions (low stress and high stress) in connection with two different feeding and rearing systems. To avoid the appearing of PSE-meat during the journey from farm to slaughterhouse, the transportation distance should be as short as possible. Great attention has to be paid to the transportation vehicle and pre-slaughter handling. During transportation the quality of the vehicle, ventilation, stocking densities and the travel distance are important factors influencing the stress-level of the pigs /1,3,4,/. Poor on farm handling increases the susceptibility to pre-slaughter stress /2/.

The carcass quality is affected by several factors, of which pre-slaughter handling in general seems to be one of the most important. Carcass quality (especially skin damages) depends on the type of rearing. The skin of partly outdoor pigs was damaged by mosquitos. Transport and pre-slaughter handling also affected carcass quality. The pigs slaughtered in Linnamäe Peekon had less skin damage. The pigs` intravitalis activity did not affect carcass quality (except mosquito'-bites). When comparing the skin damage by slaughter condition, it becomes obvious that the middle part of the carcass was less damaged in LP, but the front part and the fore shank were less damaged in SMP. Carcasses had less scratches and bone fractures in LP. The technological process in small slaughterhouses brings about a lower amount of defects. Big slaughterhouses like Saaremaa MP have a high mechanization level. In such slaughterhouses the carcass quality depends greatly on the machinery. Experimental results indicate better carcass quality of pigs slaughtered in LP.

## Conclusions

In partly out-door conditions the pigs grow faster and the amount of used feed per pig and per kg of pork is higher.

Dressing % is higher in pigs produced in partly out-door conditions and is also depending on slaughter conditions. Higher dressing % was indicated in pigs slaughtered in Linnamäe Peekon (low stress, no long transportation).

The results of the research show that transport was the most stressful procedure for pigs.

It is important to load the pigs (especially the outdoor ones) very slowly and calmly.

Rearing, feed and place of slaughter had a significant influence on the carcass temperature.

Rearing methods had great influence on the back fat thickness.

Rearing animal welfare aspects, pigs and pork characteristics, partly out-door rearing is recommended in some piggeries in Estonia.

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Scheme of production and slaughtering of pigs Partner 9 (Estonia)



CoMST 2004	
i0 <sup>th</sup> International Congress of Meat Science and Technology, Helsinki, Finl	and



Trait	Place Le M	Place Least Square Mean		Rearing Least Square Mean		P - value	Feed Lea M	ist Square ean	P - value
	LP	SLT		LP	SLT		LP	SLT	
pH45min	6,115	5,962	0,0124	6,117	5,960	0,0099	6,075	6,002	0,2269
pH 24h	5,792	5,947	0,0001	5,835	5,905	0,0003	5,900	5,840	0,0017
t° 45 min	39,082	37,192	0,0001	38,392	37,882	0,0621	38,00	38,275	0,3105
t° 24h	3,55	3,329	0,0001	2,702	4,176	0,0001	3,391	3,487	0,0409
Backfat1, mm	15,050	15,275	0,7786	13,775	16,550	0,0009	15,900	14,425	0,0684
Backfat2, mm	15,275	16,500	0,1103	14,675	17,100	0,0020	15,675	16,100	0,5766
Backfat3, mm	13,425	24,725	0,0001	12,900	25,250	0,0001	18,475	19,675	0,1746
Backfat4, mm	33,225	23,950	0,0001	31,100	26,075	0,0001	28,225	28,950	0,5168
Moisture, %	69,093	70,480	0,0004	69,653	69,919	0,4771	69,717	69,856	0,7099
Protein, %	22,262	22,430	0,4320	22,622	22,070	0,0111	22,347	22,345	0,9906
Fat, %	5,878	5,401	0,2759	5,805	5,474	0,4496	5,732	5,547	0,6707
Ash, %	1,147	1,153	0,6941	1,163	1,137	0,1118	1,139	1,160	0,2061

## Table 1. Statistical results



## EFFECT OF FREE AND RUMINALLY-PROTECTED FISH OILS ON FATTY ACID COMPOSITION, SENSORY AND OXIDATIVE CHARACTERISTICS OF BEEF LOIN MUSCLE

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#### Background

It is recommended that the concentrations of long chain polyunsaturated fatty acids (PUFA) such as eicosapentaenoic acid (C20:5*n*-3, EPA) and docosahexaenoic acid (C22:6*n*-3, DHA), for which oily fish are an excellent source, should be increased in the human diet (Simopoulos, 1998). Rumen biohydrogenation in cattle limits the amount of long chain PUFA which can be transferred into ruminant muscle, but this process also results in the production of *trans*-C18:1 and conjugated linoleic acid (CLA; particularly the trans-10, cis-12 CLA) intermediates in the rumen. It is recognised that the main sources of CLA, which may confer a health advantage to humans, are meat and milk from ruminants (Chin et al., 1992). The amount of CLA in muscles of animals given increased amounts of unprotected n-3 fatty acids was increased two- to three-fold whilst *trans*-C18:1 increased four- to seven-fold (Enser et al., 1999) with free fish oil being more effective at inhibiting rumen reductases than linseed oil.

We have previously demonstrated that feeding concentrates containing free fish oil, in comparison to megalac (a saturated fat source), increased the content of long chain n-3 PUFA in beef muscle and lowered the n-6:n-3 ratio (Scollan et al., 2001). However, the P:S ratio was unchanged, reflecting the high degree of ruminal biohydrogenation. This might be overcome by protecting the fish oils against ruminal biohydrogenation, yet some free fish oil should be available in order to encourage CLA production. The increase in long chain PUFA in beef meat had significant negative effects on colour shelf life and organoleptic properties in previous work (Vatansever et al., 2000), hence it is important to feed supra-nutritional concentrations of the antioxidant, Vitamin E, and to monitor lipid and colour oxidation and sensory aspects of the meat produced.

It is hypothesised that the use of protected fish oil, in conjunction with some unprotected fish oil, in the diet of ruminants represents a useful method for simultaneously targeting the three major objectives of producing healthy beef meat, which are (1) increase the P:S ratio, (2) reduce the n-6:n-3 ratio and (3) increase CLA content.

## Objectives

The current study was designed to manipulate the long chain PUFA and CLA content of beef fat and muscle by feeding free and ruminally-protected fish oil. The effect of the level of feeding of the protected fish oil on beef muscle fatty acid composition and its effects upon sensory characteristics and colour and lipid oxidative stability during simulated retail display were also determined.

## **Materials and Methods**

Thirty two Charolais steers with an initial mean liveweight of 619kg (s.e 7.9) were allocated on age and live weight to one of four dietary treatments, each consisting of eight animals. The 4 diets consisted of *ad libitum* grass silage plus one of four concentrates in which the lipid source was either Megalac (high in saturated palmitic acid; 16:0 from palm oil) and 100g free fish oil (FFO; rich in EPA and DHA) or megalac/FFO supplemented with a ruminally protected fish oil supplement (PFO) which was fed separately. Diet 1 megalac/FFO (control); Diet 2, megalac/FFO and 50g/d PFO (PFO1), Diet 3 megalac/FFO and 100g/d PFO (PFO2) and Diet 4 megalac/FFO and 200g/d PFO(PFO3). The PFO supplement comprised soyabean and fish oil (tuna rich in DHA), and was prepared and protected from ruminal biohydrogenation by encapsulating the lipids in a matrix of rumen inert protein (Scott et al., 1971). The PFO was considered as part of the



overall concentrate allocation per day in maintaining an overall forage:concentrate ratio of 60:40 on a DM basis. The diets were formulated so that the total dietary oil intake was approximately 5.5% (45% of which was the test oil) of total diet. Vitamin E was added to the concentrates at a level of 350IU/kg.

Animals were slaughtered conventionally and carcasses chilled overnight. At 48h post-mortem, samples of m. *longissimus thoracis* at the 11th rib were removed and blast frozen and stored at  $-80^{\circ}$ C for subsequent fatty acid analysis. Lipid was extracted using chloroform/methanol and the neutral and polar lipids separated by silicic acid column chromatography. Fatty acid methyl esters were prepared by alkaline hydrolysis followed by methylation with diazomethane and analysed on a CP Sil 88, 100m x 0.25mm ID column (Chrompack, UK). An additional 180mm section of m. *longissimus lumborum* was conditioned at 1°C for 12 days in vacuum pack. A 100mm section was then frozen at -20°C for sensory analysis. After overnight thawing at 1°C, 20mm thick steaks were cut and grilled to 74°C internal temperature. The meat was assessed by a 10 person trained taste panel using 100mm unstructured line scales. Four steaks 20mm thick were cut from the remaining sample, packed in modified atmosphere trays (O<sub>2</sub>:CO<sub>2</sub>, 75:25) and subjected to simulated retail display (700lux lighting for 16h a day, 4°C±1°C). Colour (L\*a\*b\*) was measured on the surface of two steaks at three points, daily with a Minolta Chromameter. The remaining steaks were taken at 10d of display and analysed for thiobarbituric acid reacting substances (TBARS) by the methods of Tarladgis et al. (1960) as a measure of lipid oxidation.

#### **Results and discussion**

There were no effects of diet on total DM intake, forage or concentrate intake, liveweight gain during the feeding period, carcass weight, conformation or fatness (results not shown). There were also no significant differences in total, neutral or phospholipid contents in muscle between diets (Table 2).

The 22:6*n*-3 content of muscle total lipid increased by 3.5 fold from the control to the highest level of supplementation (Table 1), all of which came from the phospholipid fraction. The content of 20:5*n*-3 also increased whilst 22:5*n*-3 decreased. CLA did not change and *cis*-18:1*n*-9 and 16:1 decreased. There were no changes in the main long chain PUFA in the neutral lipid fraction, which is very low in these fatty acids. CLA did not increase with increasing PFO, but the concentration in total lipid was similar to that found in our previous studies when free fish oil was present in the diet (Enser et al., 1999).

Total saturated fatty acids fell non-significantly by a small amount, in the PFO diets, and the total PUFA increased only slightly in the total lipid, which gave no change in the P:S ratio whether calculated by incorporating the long chain PUFA or not (Table 2). The n-6:n-3 ratio, as expected, did not change when calculated as 18:2*n*-6:18:3*n*-3, but when calculated as total n-6 PUFA to total n-3 PUFA showed a small but significant decrease with increasing PFO (Table 2). There was little change in total PUFA, the increasing EPA and DHA displacing mainly monounsaturated fatty acids, but at nearly 200mg/100g of muscle, a 100g serving would make a substantial contribution to daily requirements.

Despite the fairly small changes in PUFA content of the muscle, there were large significant effects on lipid stability. TBARS rose significantly between the control and the highest concentration of PFO in the diet. The long chain PUFA have more unsaturated double bonds and are correspondingly more susceptible to oxidation than those fatty acids with only one or two double bonds. It should also be noted that there was sufficient incorporation of EPA and DHA from the control diet, which contained free fish oil, to give a higher value for TBARS than is normally seen with this control diet (Vatansever et al., 2000). From day 7 of display it is clear that the control diet gave slightly more colour stable meat than those with PFO, but that the 7d display required by UK supermarkets is achieved by most samples (Figure 1). The highest level of PFO produced meat that was more tender than the control (Table 3), but which developed significantly higher abnormal flavours. Despite this there was no significant effect on overall acceptability.

## Conclusions

Feeding protected fish oils increased the nutritionally important long chain n-3 PUFA in muscle. Since these PUFA are predominantly deposited in phospholipids, which are diluted by the more saturated fatty acids in neutral lipids results in only small effects on the usual indices of healthy nutrition such as n-6:n-3 and P:S ratios. The incorporation of some free fish oil produced an expected concentration of CLA, which did not increase with increasing PFO, despite its imperfect protection. Long chain PUFA are sufficiently, oxidatively unstable that even small increases in their concentration made the total lipid more unstable and had a small effect on colour stability, as seen previously (Vatansever et al., 2000). Despite producing more



apparent off-flavours there was some evidence that the highest level of PFO incorporation produced more tender meat and overall hedonic acceptability was unchanged.

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Table 1. Effect of diet on composition of total fatty acids (mg/100g muscle) of m. longissimus thoracis.

	Mega	PFO1	PFO2	PFO3	s.e.d.	Р
14:0 myristic	143.3	129.2	115.3	132.1	30.60	ns
16:0 palmitic	1321	1167	1096	1189	207.4	ns
18:0 stearic	647.3	549.3	519.8	569.8	86.7	ns
18:1 <i>trans</i>	100.9	103.3	97.2	99.0	17.69	ns
18:1 <i>n</i> -9 oleic	1669	1391	1305	1486	207.1	ns
18:1 <i>cis</i> -11	72.2	62.4	57.7	63.1	7.93	ns
18:2 <i>n</i> -6 linoleic	87.8	81.3	87.0	92.9	6.67	ns
18:3 <i>n</i> -3 $\alpha$ -linolenic	24.7	23.2	23.2	25.5	1.940	ns
CLA cis-9, trans-11 C18:2	20.7	20.8	20.1	20.9	3.84	ns
20:4 <i>n</i> -6 arachidonic	23.7	22.2	24.6	24.6	1.98	ns
20:5 <i>n</i> -3 eicosapentaenoic (EPA)	$13.70^{a}$	14.09 <sup>a</sup>	14.93 <sup>a</sup>	18.39 <sup>b</sup>	1.622	0.035
22:5n-3 docosapentaenoic (DPA)	) 22.46 <sup>b</sup>	$20.71^{ab}$	19.73 <sup>a</sup>	18.61 <sup>a</sup>	1.145	0.016
22:6n-3 docosahexaenoic (DHA)	3.36 <sup>a</sup>	7.04 <sup>b</sup>	9.76 <sup>c</sup>	12.01 <sup>d</sup>	0.801	< 0.001

Table 2. Total fatty acids (mg/100g muscle), nutritional indices of total lipid of m. longissimus
thoracis and Thiobarbituric acid reacting substances (TBARS, mg malonaldehyde/Kg meat) values
of m. longissimus lumborum after 10d display in MAP

	Mega	PFO1	PFO2	PFO3	s.e.d.	р
Total fatty acids	4698	4092	3858	4258	614.2	ns
Total neutral lipids	4191	3632	3390	3785	611	ns
Total phospholipids	507	460	467	473	33	ns
18:2 <i>n</i> -6:18:3 <i>n</i> -3	3.58	3.52	3.78	3.66	0.142	ns
Total n-6:Total n-3	$1.70^{a}$	1.55 <sup>b</sup>	1.61 <sup>ab</sup>	1.56 <sup>b</sup>	0.060	0.043
P:S†	0.06	0.06	0.07	0.07	0.007	ns
P:S‡	0.107	0.118	0.131	0.128	0.015	ns
TBARS	1.64 <sup>a</sup>	2.24 <sup>a</sup>	2.33 <sup>a</sup>	3.12 <sup>b</sup>	0.361	0.004

<sup>abc</sup>Figures with the same superscript do not differ significantly

† 18:2*n*-6+18:3*n*-3:(C12+C14+C16+C18)

‡ Total PUFA:(C12+C14+C16+C18)





Figure 1. The effect of days displayed upon the change in colour saturation (±stdev) of modified atmosphere packed loin steaks, from animals fed varying levels of protected lipid supplement.

	Control	PFO1	PFO2	PFO3	sed	sig
Attributes						
Toughness	43.8 <sup>ab</sup>	45.8 <sup>b</sup>	47.4 <sup>b</sup>	38.4 <sup>a</sup>	32.81	0.05
Juiciness	$40.4^{bc}$	41.8 <sup>c</sup>	34.2 <sup>a</sup>	35.7 <sup>ab</sup>	32.65	0.05
Beef	23.7	24.7	22.3	19.8	21.68	ns
Abnormal	14.9 <sup>a</sup>	$14.0^{a}$	$15.7^{a}$	21.2 <sup>b</sup>	32.40	0.05
Greasy	13.6	12.6	14.9	14.1	01.68	ns
Bloody	3.2	7.1	4.7	4.8	21.46	ns
Livery	5.7	3.9	5.9	5.9	01.99	ns
Metallic	3.4	6.2	5.4	3.3	21.40	ns
Bitter	4.2	3.1	2.3	2.4	01.31	ns
Sweet	1.9	2.5	3.4	1.4	21.80	ns
Rancid†	1.6	1.8	2.5	3.6	10.99	ns
Fishy	1.3	1.2	2.6	3.9	21.18	ns
Acidic	9.7	11.3	9.9	7.8	02.19	ns
Cardboard	4.4	4.1	6.1	2.6	11.57	ns
Vegetable/grassy	6.2	6.1	6.1	8.4	01.85	ns
Dairy	6.9	6.1	5.7	9.0	02.27	ns
Hedonic						
Overall liking	22.5	22.3	19.2	19.1	2.42	ns

Table 3. Effect of Protected Fish Oil Supplement on sensory values of grilled loin steaks Values are the means derived from analysis of variance with Supplement and Assessor as factors; panels are treated as a 'block structure' with 8 replications.

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## INFLUENCE OF SEX, SEASON, WEIGHT AT SLAUGHTER AND MUSCLE TYPE ON BARROSÃ VEAL CHARACTERISTICS

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#### Background

The colour of fresh meat is an extremely important quality issue influencing the consumer's purchase decision. That characteristic is influenced by the pigment content (Lindahl *et al.*, 2001), the chemical state of the pigment and the microstructure of muscle tissue determined by the rate and extense of the pH fall. The relative influence of each of these factors depends on the muscle considered (Guignot *et al.*, 1992).

Barrosã calves are produced according to a traditional extensive production system in a region well known for its mountains and uplands where the winter is the main period of feed scarceness. Since they have not suffered as much genetic pressure as other breeds, the biochemical characteristics of their muscles could be different. No work was done therein to characterize the meat of Barrosã breed from the production point of view.

## Objectives

The main objective of this study was to investigate the relationship between fibre metabolic/contractile profile and some other traits veal characteristics evaluated on *Longissimus dorsi*, *Supra spinatus* and *Biceps femoris* muscles.

#### Materials and methods

Around 1 hour after slaughter, samples from *Longissimus dorsi* (Ld), *Supra Spinatus* (Ss) and *Biceps femoris* (*Bf*) of twenty Barrosã calf carcasses of both sexes, with ages ranging from 6 to 10 months were taken and frozen in liquid nitrogen for histochemical analysis as described by Roseiro *et al.* (2003). Sixty carcasses were selected for pH, colour and haem pigment analyses. Samples of the three muscles were taken and frozen at -18°C until assessment of haem pigments (Hornsey, 1956). Results were expressed as  $\mu$ g haematin/g wet tissue, using a standard curve of haematin instead of the factor used by Hornsey (1956). The pH was measured at 24h after slaughter using a glass electrode (Mettler Toledo LoT406-M6-DXK-S7/25). The colour parameters (Lab) were measured 24h *post-mortem* on the carcass surface using a Minolta CR300 in the CIE L\* a\* b\* space.

Vitamin E was determined according to European Standard-EN 12822 with minor modifications. 20g of sample trimmed of visible fat and connective tissue was weighed into a dark flask added of 1g of pyrogallic acid and 200 mL of saponification solution. This solution was prepared fresh each day and comprised 50 mL of 60% potassium hydroxide solution and 150 mL of ethanol absolute. The mixture was heated at reflux for 40 min at 100° C and then cooled and extracted 3 times with petroleum ether. After the evaporation of the solvent under vacuum, the residue was redissolved in 5 ml of hexane HPLC grade and filtrated through a membrane Acrodisc 25 mm GHP, GF 0.20  $\mu$ m (Gelman Sciences, Inc.).  $\alpha$ -Tocopherol was measured by HPLC (Spectra-Physics, model Spectra 100) using a Spherisorb S 5W silica, 5 $\mu$ m, 4.0x125 mm cartridge (Waters PSS 845549) at 292 nm. Hexane/1,4-dioxane (99:1) was used as mobile phase at a flow rate of 0.8 mL min<sup>-1</sup>. Recovery of  $\alpha$ -tocopherol from meat was determined by the addition of an internal standard to samples before saponification. Detector signals were quantified using peak areas and a calibration curve. The  $\alpha$ -tocopherol content was expressed as mg/100 g of muscle

Data were analysed using one-way and two-way analysis of variance (ANOVA) and significant differences were determined using Tukey's HSD post hoc test (Statistica 6.0-StatSoft Inc., 2001) Differences were considered different at p < 0.05.



#### **Results and discussion**

#### Muscle traits

Least square means of fibre type composition, colour parameters, pH<sub>24</sub>,  $\alpha$ -tocoherol and haem pigment concentration obtained from *Longissimus dorsi*, *Biceps femoris* and *Supra spinatus* muscles are presented in Table 1. Significant differences were found among muscles for all traits studied except for b\* value and  $\beta R$  fibres percentage. Ss muscle presented higher mean  $\alpha$ -tocopherol (P<0.05) and haem pigment (P<0.001) than Ld and Bf muscles, which did not differed. Regarding pH<sub>24</sub>, Ss and Ld muscles showed significantly higher mean values than Bf muscle. There were no significant differences between Ld and Bf muscles in regarding to fibre metabolic and contractile profiles, except for  $\alpha W$  fibres. Bf muscle showed the highest frequency of  $\alpha W$  fibres, but not presented the highest glycolytic profile which belong to Ld. Ss muscle presented a metabolic profile significantly less glycolytic than Ld, but did not differ from Bf.

Guignot *et al* (1992) reported a contractile profile in Friesen-Holstein calves, similar results to those observed in this study, for the same muscle.

Table 1 - Compositional characteristics of Longissimus dorsi, Biceps femoris and Supra spinatus from Barrosã veal.

-	Ld	Bf	Ss	F value
$\alpha$ -tocopherol (mg.g <sup>-1</sup> )	0.172 <sup>b</sup>	0,165 <sup>b</sup>	0,216 <sup>a</sup>	4,049*
Haem pigment (mg.100g <sup>-1</sup> )	16.36 <sup>b</sup>	16,66 <sup>b</sup>	19,69 <sup>a</sup>	24,023***
PH <sub>24</sub>	5.67 <sup>a</sup>	5,46 <sup>b</sup>	5,72 <sup>a</sup>	23,50***
L*	35.22 <sup>b</sup>	39,51 <sup>a</sup>	ND	44,83***
a*	11.69 <sup>b</sup>	13,73 <sup>a</sup>	ND	49,37***
b*	2.76	2,53	ND	1,54 <sup>ns</sup>
βR fibres (%)	25.35	22,35	24,27	0,65 <sup>ns</sup>
αR fibres (%)	22.89 <sup>ab</sup>	17,56 <sup>b</sup>	26,81 <sup>a</sup>	8,98***
$\alpha W$ fibres (%)	51.76 <sup>b</sup>	60,10 <sup>a</sup>	48,93 <sup>b</sup>	6,21**
Oxidative fibres (%)	48.52 <sup>b</sup>	52,20 <sup>ab</sup>	53,96 <sup>a</sup>	4,55*
Glycolytic fibres (%)	51.48 <sup>a</sup>	47,80 <sup>ab</sup>	46,04 <sup>b</sup>	4,55*

In same row, means with different letters are significantly different. \* P<0.05; \*\* P< 0.01; \*\*\*P<0.00;

ns-not significant; ND-not determined; L\*,a\*,b\*- colour parameters.

The number of fibres in bovine muscles is almost determined at birth. However, the relative fibre type composition is affected to a number of genetic and environmental factors such as age, sex, breed and exercise performance. The main fibre type change occurs in the first few months of life. Based on different breeds, which varied in age between 0 and 12 months, Wegner *et al.* (2000) stated that such fibre type conversion occurred as early as 6 months old and was characterized by an increase and decrease frequency of type IIb and type IIa fibres, respectively, and no alterations in the type I incidence.

All Barrosã calves used in our study were more than 6 months old and the age effect on muscle fibre profile was not evaluated. Nevertheless, there was a remarkable effect of the weight at slaughter on contractile (P<0.001) metabolism (Table 2). Attending to the low correlation between the animal age and weight at slaughter, that influence could be attributed to differences in diet and animal handling. Listrat *et al* (1999) referred that muscle fibre characteristics could be related to the energy level of the diet and to daily dry matter intake. However, the diet effect on muscle fibre type profile of calves are not easily explained since during the most important period for fibre type changes, they are basically fed with mother's milk. The impact of diet on muscle fibre profile of calves reared in an extensive system needs more attention.

The influence of sex on meat traits was generally less expressive inducing significant changes only on  $\beta R$  and  $\alpha W$  fibres and  $pH_{24}$ . Dreyer *et al.* (1977) reported that muscles of bulls contained more red fibres than those of steers. Johnston *et al.* (1981) found a significant higher percentage of  $\alpha W$  fibres in Ld muscle of heifers than in steers. In our studied population, male muscles showed significantly (P<0.001) higher and lower percentages of  $\beta R$  and  $\alpha W$  fibres, respectively, than females (data not shown).



		F VALUE	
-	Weight	Sex	Season
α-tocopherol	0,49 <sup>ns</sup>	0,006 <sup>ns</sup>	2,79 <sup> ns</sup>
Haem pigment	14,03***	0,519 <sup>ns</sup>	4,01*
PH <sub>24</sub>	0,40 <sup>ns</sup>	3,90*	10,70**
L*	1,81 <sup>ns</sup>	0,90 <sup>ns</sup>	1,23 <sup>ns</sup>
a*	0,22 <sup>ns</sup>	0,80 <sup>ns</sup>	0,03 <sup>ns</sup>
b*	6,04*	1,34 <sup>ns</sup>	4,39*
βR fibres (%)	7,97**	5,24*	ND
αR fibres (%)	11,77***	2,68 <sup>ns</sup>	ND
αW fibres (%)	24,44***	9,39**	ND
Oxidative fibres (%)	2,77 <sup>ns</sup>	0,47 <sup>ns</sup>	ND
Glycolytic fibres (%)	$2,77^{\text{ns}}$	0,47 <sup>ns</sup>	ND

 Table 2 – Effect of weight, sex and slaughter season on muscle characteristics.

In same row, means with different letters are significantly different. \* P<0.05; \*\* P<0.0; \*\*\*P<0.001; ns-not significant. ND-not determined; L\*,a\*,b\*- colour parameters

#### Relations between pigment content, muscle traits and production factors

Correlations between the haem pigment concentration and the other Barrosã meat parameters are depicted on Table 3, and were not significant.

The highest haem pigment content was found, as expected, in the most oxidative muscle corroborating the trend observed by Meynier & Gandemer (1991). This parameter was affected by the animal weight at slaughter (P<0.001) and the slaughter season (P<0.05) but not by sex. Irrespective of the carcass weight, the highest pigment value was measured in older animals (data not shown).

	Table 3 – Correlations between muscular traits.							
	1	2	3	4	5			
1 α-tocopherol	1,00							
2 Haem pigment	-0,23 <sup>ns</sup>	1,00						
3 pH <sub>24</sub>	-0,27 <sup>ns</sup>	0,10 <sup>ns</sup>	1,00					
4 L*	0,24 <sup>ns</sup>	-0,27 <sup>ns</sup>	-0,36*	1,00				
5 a*	-0,07 <sup>ns</sup>	0,29 <sup>ns</sup>	-0,29*	0,12 <sup>ns</sup>	1,00			
6 b*	0,23 <sup>ns</sup>	-0,11 <sup>ns</sup>	-0,52***	0,44**	0,51***			

\* P<0.05; \*\* P< 0.01; \*\*\*P<0.00; ns-not significant. L\*,a\*,b\*- colour parameters.

The effect of slaughter season on haem pigment content as well as on  $pH_{24}$  is not easily comprehensible, but it could possibly be related to animal handling, diet composition and feed availability. Although animals are slaughtered throughout the year, spring and autumn can be described as stronger production seasons. The grass availability and its composition are not regular all over the year and this fact could have implications in growth traits and influence the carcass and meat quality. The pigment concentration referred in our study for Ld muscle was similar to those reported by Gil *et al.* (2001), in older animals of Retinta breed.

#### Relations between colour, pH, muscle traits and production factors

Due to its difficult access, colour parameters were not measured on Ss muscle. Comparing the other two muscles, L\* and a\* values were significantly higher in Bf than in Ld muscle (P<0.001). The absence of a significant correlation with the pigment content confirms the results obtained by Hunt & Hedrick (1977) and Tam *et al.* (1998) and disagreed with the findings of Lindahl *et al.* (2001). Although McDougall (1982) claimed that the pigment content was the main factor affecting muscle colour, that relationship depended on the muscle location (Guignot *et al.*, 1992).

A study of Priolo *et al.* (2001) have shown that meat from animals finished on pasture is darker than those finished on concentrate, with the formers presenting a L value about 5% lower. In accordance to Page *et al.* (2001) a significant correlation between muscle  $pH_{24}$  and colour values was also observed. This correlation can be explained in that colour in muscle tissue is based on the reflectance of light off free water and on



oxygenation of the myoglobin At a higher muscle pH, proteins are bound strongly with water, allowing less free water to escape to interstitial space around cells and the muscle fibres to swell. Therefore, meat with higher pH will be darker in colour because there is less free water to reflect light. These authors also stated that a higher  $pH_{24}$  produced less red and yellow beef. Our results agreed with Page's experiment. The Ld muscle showed a significant higher mean  $pH_{24}$ value (P<0.001) and lower L\* and a\* values (P<0.001) than Bf muscle.

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# IMPORTED AUSSIE BEEF QUALITY AND COMPARISON WITH US BEEF

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#### Background

Meat consumption in Japan is considerable at present, with beef imports particularly high from the USA and Australia. Certain brands are quite popular and have become standards of choice. But this merely reflects personal preferences and attitudes, without basis in objective data. The Japanese domestic beef (black cattle) is of course best known to the people in Japan. The breeding periods and types of feed differ for this domestic brand. But little research has been conducted of the meat quality of imported beef.

#### Objectives

Consequently, the present study was conducted to address this matter using Aussie beef frequently to be found in stores in Japan. Based on the results, comparison is made with American brand (US) beef considered as equivalent in quality to or even better than the former.

#### Materials and methods

<u>Beef sample:</u> Strip loin samples of Aussie and US beef (N=4) were chilled and aged for 30 days for sensory and physicochemical analysis. All samples of either brand were graded as Choice or better. The Australian species, Angus cross, is primarily fed barley while the American Angus cross is given feed whose major constituent is corn.

<u>Sensory evaluation</u>: Evaluation in all cases was made using beef (thigh side) sliced at 8 mm in thickness. Raw meat and meat grilled for 2 min, (1 min for each side) at 180°C by an electric plate were evaluated by a team of experts from Nippon Meat Packers using the two-point comparison method.

<u>Color of meat and fat</u>: Lab-values were obtained for lean and fat of the 8 mm slices using a color difference meter (Minolta CR-200). Lean color was assessed after 2 days store-front storage under fluorescent lighting for meat packed with plastic  $O_2$ -permeable film at 4°C.

<u>Myoglobin content:</u> Beef samples were cut up into pieces for homogenization and myoglobin (Mb) content as well as the met-Mb (MetMb) ratio in minced meat were determined by the extraction method (Sakata and Nagata, 1992; Trout, 2003).

<u>TBA value</u>: Distillation was carried out for determination of malonaldehyde (MA, Yamauchi and Ando, 1973) in the above minced meat sample. TBA value was expressed as MA mg/1000g meat.

<u>Water and fat contents</u>: Using minced meat, water and fat contents were determined by the freeze-dried and Soxhlet extraction methods, respectively.

<u>Water holding capacity</u>: Five cm thick beef cuts were vacuum-packed and cooked at 70°C for 90 min. Cooked meat yield (%) of meat following water loss due to cooking was designated as water holding capacity. Using a press-machine, beef  $(10 \times 10 \times 5 \text{ mm})$  was pressed at  $15 \text{kgw/cm}^2$  for 40 sec. The amount of water released and pressed meat area were determined based on the water holding capacity of raw meat.

<u>Hardness assessment</u>: Beef hardness of the 8 mm slices was assessed by the multi-bite method using a Tensipressor (Taketomo TTP-50BXII).

<u>Melting point and area % of muscular fat:</u> Muscular fat extracted by the Soxhlet method was used to determine fat melting point using apparatus specifically designed for this purpose.

<u>Ratio of fat size in lean muscle:</u> The 8 mm meat slices were photographed and monochrome figures were incorporated into computer provided with an image analysis system (Pias LA525). Using this system, fat marbling and lean portions were divided into black or white sample groups and on the basis of the size ratio, fat size (% of whole meat area) and number were determined.



<u>Composition of fatty and amino acids in lean muscle:</u> Fatty acid composition was clarified by gas - chromatography (Hewlett-Packard 6890) using muscle specimens. Amino acid analysis on minced beef was carried out for clarification of free and peptide-forms using an amino acid analyzer (JEOL JLC-500/V) and carnosine were determined at the same time.

#### **Results and discussion**

<u>Sensory evaluation</u>: Table 1 shows the results for sensory evaluation of the 2 beef brands. Aussie raw beef exhibited a darker red and the fat was white, in obvious contrast to the US beef. The grilled samples were assessed as better aroma, more tasty and more tender. But these qualities differed according to the sample. US beef was generally considered more tender.

<u>Color of meat and fat:</u> Table 2 shows changes in color with storage. On 0 day (1 hr after slicing), the L value was greater in US beef, with a and b values essentially the same. This would account for its brighter red color. On day 2, L had decreased and a value was greater. Changes in Aussie beef were fewer during this period. US beef thus has greater tendency to undergo discoloration. Table 3 indicates fat color for the 2 brands and L to be higher and a lower in Aussie beef. It thus follows that Aussie beef has greater white fat content.

<u>Myoglobin content:</u> Mb content and MetMb% during storage are shown in Table 4. Mb content was greater in Aussie beef. At 2 days storage, Mb content of both brands had decreased, suggesting the possibility of drip from meat surface during storage. MetMb increased with the period of display, more so in US than Aussie beef.

<u>TBA value and carnosine content:</u> Tables 5 and 6 show respectively values for TBA and carnosine. TBA value was higher in US beef on day 0 and 2 while Aussie beef contained greater carnosine.

<u>Water content</u>, cooking yield and water holding capacity: Table 7 shows water content for lean meat and Table 8, water holding capacity (WHC), The values were basically the same for the two brands in cooked meat yield (Table 8-1), released water (-2) and area of pressed meat (-3).

<u>Hardness assessment</u>: Table 9 shows the value of this parameter as determined using a Tensipressor. The US beef was generally significantly more tender.

<u>Fat content in lean muscle and melting point:</u> Table 10 shows fat content in lean muscle and its melting point. The values were essentially the same for the 2 brands. Aussie beef had greater fat content (Table 11), this being consistent with greater degree of fat marbling as seen on the computer image display. US beef exhibited significantly lower fat melting point.

<u>Fat and amino acid composition:</u> Myristoleic acid (C14:1) and arachidic acid (C20:1) were higher in US beef and stearic acid (C18:0) was significantly abundant in Aussie beef. Ratios of unsaturated fatty acids were significantly lower in Aussie beef, this possibly causing higher fat melting point. Glutamic acid was higher, through not significantly, in Aussie beef (data not shown).

The results for sensory evaluation and color measurement of raw beef indicated Aussie beef fat to be lighter in color and meat color darker red compared to US beef. This reflects the chromatic character of Aussie beef, which is superior in quality owing to its fatness character. US beef appears to incur more discoloration owing to fluorescent lighting in the display case, while that of the former is stable maintained. The MetMb ratio was higher in US beef subsequent to display, causing the meat to become brownish. TBA was lower in Aussie beef with consequently less rancidity. This feature and discoloration are due to oxidation which has been shown to occur less in Aussie beef. Aussie beef capacity for preservation is thus greater. The presence of relatively more carnosine, an antioxidant, may be one reason for this. The sensory evaluation of grilled beef, aroma, taste and tenderness showed essentially the same results for the two brands. The evaluation team gave the same scores for eating quality. US beef was considered a little more tender than the Aussie brand. No relationships could be detected among tenderness, water holding capacity and fat marbling. US beef in this study showed looser muscle fibers and this possibly may explain its somewhat greater tenderness.



## Conclusions

Based on the results of this study, the following conclusions were drawn: Subcutaneous fat in Australian meat cattle was at one time noted to be almost entirely yellow with little marbling. This was due to the cattle feeding mainly on grass. But grain feed with special formula for fattening has resulted in improved meat quality and greater meat exports to Japan. Aussie beef is shown by the present study to be superior in quality owing to its greater water holding capacity. The Japanese are grateful for this high quality beef. Aussie beef imports will continue to increase until BSE inspections in the USA is fully implemented with satisfactory results.

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Table 1. Results of sensory evaluation of beer							
		Aus. beef	US beef	No difference			
Raw	Darker red (meat)	67	26	8***			
	More white (fat)	88	7	6***			
	Greater aroma	39	53	9			
Cooked	Greater aroma	41	55	5			
	More tasty	41	55	5			
	More tender	34	63	4*			
	More delicious	45	41	5			

1 ... T 1 1 D 14 C C1 0

Figures indicate numbers of evaluation personnel.

Showed significant differences at p<0.05 (\*) and p<0.001 (\*\*\*).

Table 2	Changes	in	boof	aalar	with	storage
1 a O C 2.	Changes	ш	UCCI	COIOI	with	Storage

	0 d	0 day		2 days		Aus. beef US be	
	Aus. beef	US beef	Aus. beef	US beef	L	74.27	$70.70^*$
L	38.74	40.73*	38.46	38.77	а	7.46	9.12*
а	20.11	20.75	20.80	19.72 <sup>*</sup>	b	9.00	9.18
b	6.56	6.90	6.39	7.20	*	* n<0.05	
						P	

Table 3 Fat color of beef

\* p<0.05

Table 4.	Mb	content	(mg/	g meat)	and	MetMb	(%).
	1.10	•••••••	0	8		1.1.0.1.10	(, , , , , , , , , , , , , , , , , , ,

Mb content (mg/g meat)			MetMb (%)				
0 d	ay	2 da	iys	0 d	ay	2 da	iys
Aus. beef	US beef	Aus. beef	US beef	Aus. beef	US beef	Aus. beef	US beef
7.15	6.33*	6.72	5.83*	8.71	6.55	39.48	45.89**

\* p<0.05, \*\* p<0.01

Table 5. TBA values during storage

0 0	day	2 days		
Aus. beef	US beef	Aus. beef	US beef	
0.342	1.390*	0.767	$2.422^{*}$	
* p<0.05				

Table 6. Carnosine content (umoles/g meat	Table 6.	Carnosine content	(umoles/g meat)
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Aus. beef	US beef
26.5	22.14*
* p<0.05	

Table 7. Wat	er content (%)
Aus. beef	US beef
69.60	69.08

Aus. beef	US beef
78.92	79.88

Table 8-2.	WHC:	Released	water	(%)
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# Table 8-3. WHC: Area of pressed meat (cm<sup>2</sup>/g meat)

Aus. beef	US beef
23.07	24.65

	Č	<u> </u>
Aus. beef	US beef	
21.27	21.92	

Table 9.	Hardness	$(kgw/m^2)$
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Aus. beef	US beef
24.73	22.67*

<sup>\*</sup> p<0.05

Table 10. Muscular fat content and its melting point

		U	1				
Fat conte	nt (%)	Melting point (°C)					
Aus. beef	US beef	Aus. beef	US beef				
6.81	6.88	41.5	$40.2^{*}$				
* -0.05							

\* p<0.05

Fat siz	æ (%)	Fat nu	mber
Aus. beef	US beef	Aus. beef	US beef
10.03	6.85	155	182

Measured by computer provided with image analysis system.



# THE EFFECT OF REFRIGERATION CONDITIONS ON THE QUALITY OF LAMB MEAT

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#### Background

In Portugal the traditional lamb meat production systems are based on local breeds, raised on pasture for most of the year. Many of these products are protected by the EU with denominations of origin (PDO) or geographic indications (PGI). Consumer's normally consider certified meat as healthy food, produced in natural conditions and have high expectations concerning eating satisfaction. Certified meat is marketed at higher prices than undifferentiated meat and is required by high quality segments of the market. Particularly for these products, it is essential to control all the production processes in order to obtain the highest quality standards and to reduce variability. In Portugal, in spite of being recommended for several lambs with PDO or PGI the practice of low carcass cooling rates, the common practice is to cool carcasses as soon as possible in order to reduce evaporative weight loss and prevent spoilage.

Tenderness is one of the most important organoleptic characteristics, and only tender meat can assure consumer's satisfaction. The main factors affecting tenderness are sarcomere length, proteolysis of myofibrillar proteins and connective tissue (Koohmaraie *et al.* 2002). The rates of temperature and pH decline in the early *post mortem* play an important role in meat tenderness. A slow cooling rate before the onset of *rigor mortis* prevent cold shortening and stimulate the proteolysis of myofibrils and associated proteins (Lockner *et al.*, 1980; May *et al.*, 1992), increasing meat tenderness. The effect of rapid cooling of carcasses may be particularly important in small carcasses, such as lamb, and Geesink *et al.* (2000), in a trial with lamb *longissimus* muscle concluded that optimal temperature at the onset of *rigor mortis* is around 15 °C. In these conditions muscle shortening was minimized and proteolysis during storage was unaffected.

#### **Objectives**

The objective of this trial was to evaluate if 4 and 8 hours at 12 °C, before cooling lamb carcasses at 2° C affects tenderness, histological, biochemical, physical and microbiological traits of meat.

#### Materials and methods

#### Animals and sampling

Thirty Merino Branco ram lamb carcasses were used to conduct this trial. Lambs were slaughtered with four months of age in the abattoir of the Estação Zootécnica Nacional. After dressing and weighing, three groups of ten carcasses were assigned to three refrigeration conditions: 0 (cooling at 2 °C immediately after slaughter), 4 (cooling at 2 °C after four hours at 12 °C) and 8 (cooling at 2 °C after eight hours at 12 °C). Temperature and pH of the *longissimus lumborum* (LL) and *semimembranosus* (SM) muscles were measured 4, 8 and 24 hours after slaughter, using a penetration probe electrode. Eight hours after slaughter, and at the third, seventh and tenth days, sampling to microbiological analysis were performed on carcass shoulder region by destructive method (2,5cm<sup>2</sup>) according to the Directive 64/433/CEE. Three days after slaughter, carcasses were split in two halves and the muscles LL and SM of the right sides were vacuum packed and frozen at – 20 °C until shear force determinations. The colour of *longissimus thoracis* (LT) was estimated in the L\*, a\* and b\* system (Minolta CR-300 chromometer) at the level of the 13th dorsal vertebra, after one hour of exposition to the air. Samples of LT and SM were collected, vacuum packed and frozen at – 80 °C to determine sarcomere length. A small portion of LT muscle was collected to determine water-holding capacity (WHC). In the left halves of the carcasses, LL was vacuum packed and frozen at – 20 °C until sensory analysis.

#### Analytical procedures

Sarcomere length was determined by the method described by Sañudo *et al.* (2003). Water holding capacity was determined by the method proposed by Santos-Silva *et al.* (2002).



Concerning microbiological analysis, total psycrotrophic aerobic counts at 7°C for 10 days (Plate Count Agar, Merck, Germany), *Enterobactereaceae* on VRBD agar (Merck, Germany) for 48h at 37°C, *Escherichia.coli* at 44,5°C in Tergitol BCIG agar (Biokar Diagnostics) for 24h, lactic acid bacteria counts on Man Rogosa Sharpe Agar (Oxoid, UK) incubated at 30°C for 3 days and *Brochothrix termosphacta* count in streptomycin, actidione, thallous acetate agar (STAA, Oxoid, UK) incubated for 2 days at 30°C. Counts were expressed as log cfu/cm<sup>2</sup>.

For shear force determination, samples were thawed for 24 hours at 2°C and cooked in an electric oven until the meat internal temperature reached  $65 \pm 5$  °C. Cores with a section of about 1 cm<sup>2</sup> were prepared in the muscle fibres direction. Shear force was determined using a texture analyser (TA-XT2i Texture Analyser, Stable Micro Systems) equipped with a Warner-Bratzler shear device and data were collected with specific software (Texture Expert Exceed, Stable Micro Systems). Measurements were recorded as the average value of a minimum of six replicates. For sensory analysis a trained panel of six members was used to compare tenderness of meat of the three experimental groups using a structured scale of ten points (0 – very tough; 10 – very tender). Meat was prepared as described for shear force determination. Three cores corresponding to the three cooling conditions were presented to the panellists simultaneously and served in hot plates in a total of 51 comparisons.

The data were analysed using the GLM procedure of SAS (1989). When the F-test was significant, the leastsquares means were compared. For shear force, muscle temperature, pH and sarcomere length, muscle and refrigeration conditions were used as fix effects and cold carcass weight (CCW) as covariate. For muscle colour parameters and WHC, the refrigeration condition was the fix effect and CCW the covariate. For tenderness and microbiological traits both models included the refrigeration conditions, besides panellist (tenderness) and days after slaughter (microbiological traits).

## **Results and discussion**

Shear force was higher (p<0.05) when carcasses were immediately cooled (group 0) and no significant differences were found between groups 4 and 8. Data for LL are in accordance to those obtained for sensory evaluation, as panellists considered meat of group 0 the tougher and of group 4 the most tender. These results agree with those of Marques de Almeida *et al.* (2003), suggesting that 3 or 4 hours delay of carcass cooling at 2 ° C are enough to improve meat tenderness 3 days after slaughter. Jaime *et al.* (1993), comparing the effects of 3 cooling conditions on tenderness of lamb *longissimus* muscle, concluded that toughness 2 days after slaughter was higher in carcasses with faster temperature decline, although no differences were found after eight days of ageing. Thatcher and Gaunt (1992) observed higher shear force of meat from lambs chilled at 2° C compared to others chilled at 10 °C.

As expected, the rate of temperature decline was higher, intermediate and lower for groups 0, 4 and 8 respectively, and in LL than in SM. These results for temperature explain the differences in the rates of pH decline, which was higher in groups 8 and 4 than in group 0 and in SM than in LD.

When muscle temperature reaches values below 10 °C and pH is still above 6, cold shortening may occur. The values observed for pH and temperature could suggest a higher muscle shortening, particularly in group 0. However, the values of sarcomere length indicate that shortening was very moderate in the 3 groups and no differences were found between the three refrigeration conditions or the two muscles. The correlation coefficient between shear force and sarcomere length was low (r=-0.06) and not significant (p>0.05). Marsh and Leet (1966) demonstrated that up to 20 % of muscle shortening there is no effect in tenderness of muscle *sternomandibularis*.

Shear force values showed correlations coefficients of -0.40 (p<0.05) and -0.42 (p<0.05) with muscle temperature 4 hours after slaughter in LL and SM, respectively. Other authors have reported that temperature in the initial period after slaughter is the factor most highly correlated with tenderness (Lockner *et al.*, 1980; May *et al.*, 1992), which is probably related to a higher extension of proteolysis by endogenous peptidases (Yates *et al.*, 1983). Although we do not dispose of data to confirm this hypothesis, the differences in *in situ* proteolytic activity may explain the results of this trial.

In the range of pH values above 5.5, meat with higher pH may have higher WHC (Purchas, 1990) and sow lower values of L\* (Priolo *et al.*, 2001). Also, higher rates of glycolysis, associated to lower rates of temperature decline, may affect negatively meat WHC (Hood and Joseph, 1985). In this trial the refrigeration conditions had no effect on LT colour and WHC. Thatcher and Gaunt (1992), also found minor effects on meat colour of ewe lambs chilled at 2 or 10 °C.



The results for microbiological traits are presented in table 2. The counts on the day of slaughter (0) for total psycrotrofic bacteria ( $\pm$  3 log cfu/cm<sup>2</sup>) and the results obtained for *E. coli*, that was not detected at day 0 and showed low values during conditioning, indicate good hygiene in the abattoir. On day 0 the dominant Gram+ flora seems to be lactic bacteria. Only for this group of microbes, the counts were higher (p<0,001) when temperature decline was lower (groups 4 and 8) and this effect was still observed on day 3. However, on the 7th and 10th day the results were similar for the 3 refrigeration conditions, suggesting that the increase of population reached a stationary state corresponding to values of 3-4log cfu/cm<sup>2</sup>. After the 7th day, the dominant flora is *Brochothrix thermosphacta* and the Gram– psycrotrophic bacteria. These populations are responsible for the spoilage of meat that is already evident at the 10th day, when the counts for total psycrotrophic are higher than 8 log cfu/cm<sup>2</sup>, independently of the refrigeration conditions. The shelf life period, in the conditions of slaughter and refrigeration of this trial, was up to 7 days.

## Conclusions

According to the results of this trial the rapid cooling of lamb carcasses results in tougher meat. The 4 hours delay in carcass cooling at 2 °C, seems to improve tenderness, and have no negative effects on the physical traits of meat (colour and water-holding capacity). Although the delay of cooling increased the counts of lactic acid bacteria at the day of slaughter, it did not affect the hygienic conditions of meat after 7 days. In the conditions of slaughter and refrigeration of this trial, the meat microbial shelf life period was up to 7 days.

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Table 1.	Meat	quality	traits	of	longissimus	and	semimembranosus	muscles	of	lamb	carcasses	submitted	to
three ref	rigerat	ion conc	litions	5									

		longissimus			imembrano.	SEM		Effect	S	
	0	4	8	0	4	8	-	М	G	M*G
Shear force (kg)	6.65 b	4.51 a	5.97 ab	7.18 b	5.82 a	5.77 a	0.552	ns	*	ns
Tenderness	5.81 a	7.32 c	6.67 b				0.231		***	
Temperature 4h (°C)	6.7 a	13.5 c	13.5 c	9.6 b	15.5 d	16.6 e	0.21	***	***	**
Temperature 8h (°C)	4.0 a	5.6 c	11.8 e	5.0 b	6.6 d	12.4 f	0.22	***	***	**
Temperature 24h (°C)	2.9 a	2.9 a	3.9 b	2.6 a	2.8 a	3.6 ab	0.33	*	ns	***
pH 4h	6.70	6.50	6.67	6.46	6.26	6.46	0.053	***	***	ns
pH 8h	5.98	5.97	5.76	5.97	5.87	5.76	0.059	**	ns	ns
pH 24h	5.72 b	5.69 ab	5.61 ab	5.71 b	5.65 ab	5.58 a	0.038	*	ns	*
L*	41.5	42.7	40.1				0.74		ns	
a*	11.7	11.8	12.4				0.34		ns	
b*	3.34	3.79	3.43				0.244		ns	
Water-holding capacity (%)	30.0	26.4	30.6				1.11		ns	
Sarcomere length $(\mu)$	1.99	2.09	1.96	1.69	1.72	1.76	0.047	***	ns	ns

SEM- standard error of means; M - muscle; G - group

Table 2. Effects of	f refrigeration c	conditions and	l days after	slaughter	on microbial	proliferation at	the surface
of lamb carcasses (	$(cfu/cm^2)$			-		^	

Group			0				4			1	8		SEM		Effec	ets
Days pm	0	3	7	10	0	3	7	10	0	3	7	10		G	D	G*D
Bt	a 0.308	a 0.100	cde 5.008	cde 5.023	a 0.290	b 1.262	cde 4.917	e 6.247	a 0.000	b 1.747	с 4.364	de 5.657	0.2957	*	***	**
Ent	ab 0.715	ab 0.755	de 2.873	e 3.270	a 0.460	bc 1.648	cd 2.306	f 4.474	ab 0.742	a .580	de 3.139	de 3.144	0.3387	ns	***	*
Тр	3.377	3.887	6.501	8.250	3.265	4.248	6.163	8.997	3.579	4.381	6.032	8.094	0.3138	ns	***	ns
E. coli	0.000	0.230	0.396	0.390	0.000	0.278	0.360	0.360	0.000	0.130	0.130	0.317	0.0838	ns	ns	ns
Lb	0.341	0.818	3.359	3.568	1.221	1.856	3.880	4.518	1.084	1.611	3.955	3.612	0.1326	***	***	ns

SEM - standard error of means; G - group; D - days after slaughter; *Bt* - *Brochothrix termosphacta; Ent* - *Enterobactereaceae;* Tp - Total psicrotrophic; *E. coli* - *Escherichia coli;* Lb - Lactic bacteria



## CARCASS QUALITY OF SEVERAL EUROPEAN CATTLE BREEDS: PRELIMINARY RESULTS

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#### Background

Selective breeding in cattle has been very successful in increasing production indicators, but until now there is little information and subsequently limited knowledge to allow selection programs to be designed to improve product quality. Identification of the genes involved in aspects of carcass and meat quality and the quantification of their effects will provide the basic information to devise breeding programmes that enhance quality and take account of regional variations in preference.

The present study is part of an ongoing EU project, which will examine a total of 450 animals from 15 different breeds representing the genetic diversity of European cattle. This project will undertake a rigorous comparison of carcass and meat quality from this range of cattle breeds with animals raised under similar management conditions in order to minimise environmental variation and to identify and define genetically determined component of that variation.

## Objectives

To assess the variability among 15 different European cattle breeds in carcass traits.

#### Materials and methods

The material consisted of 337 young entire males from 15 different European cattle breeds. The breeds used were as follows: From United Kingdom, Jersey, South Devon, Aberdeen Angus and Highland; from Denmark, Holstein, Danish Red Cattle and Simmental; from Spain, Asturiana de los Valles, Asturiana de la Montaña, Avileña-Negra Ibérica and Pirenaica; from Italy, Piemontese and Marchigiana and from France, Limousin and Charolais. All animals were intact bulls, which were fed *ad libitum* with a standardised diet composed of high barley proportion (about 80%), soya (9%) and chopped straw (10%) with appropriate minerals and vitamins. Energy density was approximately of 12.5 kJ/Kg dry matter.

The welfare regulations were taken into account when handling the animals. The bulls were slaughtered at the nearest EU licensed abattoir, to minimise the effect of the transport stress on meat quality. Slaughter was at 75% of mature bull weight, which in most cases was about 15 months of age. Stunning was by captive bolt pistol. Carcass dressing was carried out using a standardised project protocol that corresponded to commercial practice. Carcasses were chilled at  $4 \pm 1^{\circ}$  C for 24 hours.

Immediately after slaughter, the following variables were recorded:

- Kidney fat weight.
- Hot carcass weight, measured without removing the subcutaneous fat.
- Dressing percentage, calculated according to the following formula: 100\*hot carcass weight/slaughter live weight.
- Conformation score was graded according to the SEUROP classification (R. (CEE) 1026/91, R. (CEE) 2237/91 and R. (CEE) 2930/81) with a scale ranging from 1 (very bad conformation) to 18 (very good conformation).



- Fatness score was measured by UE classification, with a 15 points scale (1, very low fat to 15, very high fat).

Also, several standard measurements were taken on the left half carcass to evaluate carcass morphology, according to the methodology described by De Boer, Dumont, Pomeroy & Weniger (1974). Variables recorded were carcass length, internal depth of breast, limb length and limb thickness. The blockiness index was calculated. This index expresses the relationship between carcass weight (kg) and carcass length (cm). High values indicate high muscular development (Albertí *et al.*, 2001). Limb index expresses the relationship between limb thickness (cm) and limb length (cm).

The area of the *Longissimus thoracis* (LT) muscle, at 5<sup>th</sup>-6<sup>th</sup> rib level, was recorded by tracing: an acetate sheet was placed on the surface of the loin, and the border of the muscle marked on the sheet using glass marker. The area of the muscle was calculated by planimetry. Medium-lateral and dorso-ventral diameters (A and B, respectively), were also measured (Cañeque & Sañudo, 2000).

The 6<sup>th</sup> thoracic rib joint was collected at 24 h post-mortem, and its' weight was recorded together with the *Longissimus thoracis* muscle, which was separated for instrumental analysis. The rest of the rib joint was dissected. Tissue composition for muscle, bone, fat and other components (tendons and noticeable blood vessels) were estimated from the rib joint according to the method described by Robelin & Geay (1975). Results are expressed as percentage of the entire rib weight.

Statistical analysis was performed using SPSS 11.0 software. An ANOVA procedure was carried out with breed as an unique effect.

#### **Results and discussion**

Global results are shown in Table 1.

Breed was a very important factor determining carcass quality, especially in carcass blockiness index (F=96.7), carcass weight (F=96.5), dressing percentage (F= 93.2), fatness score (F= 86.6) and limb length (F=84.8). Carcass weights ranged from 189.9 kg for Jersey to 386.6 kg for Charolais. In general, dairy breeds, such as Jersey, Holstein or Danish Cattle, or local breeds, such as Highland or Asturiana de la Montaña, produced small carcasses, while specialised beef breeds, as Charolais or Pirenaica produced higher carcass weights. Similarly, dressing percentage was lowest for Jersey (46.6%) while five specialised beef breeds (Charolais, Pirenaica, Asturiana de los Valles, Piemontese and Limousin) had values up to 60%. The breed purpose was also reflected in conformation scores, the highest score was for Piemontese (14.6) and the lowest for Jersey (4.4). On the other hand, dairy and local breeds showed the highest values for kidney fat weight, which was maximum for Highland (9.2 kg) and minimum for both Italian breeds (less than 1.5 kg). Nevertheless, fatness scores did not follow this behaviour, since Charolais or Limousin had the same fatness scores as some dairy or double purpose breeds such as Simmental, Holstein or Red Danish Cattle (about 8 points). The breed with the highest fatness score was the Aberdeen Angus (11.7) and at the lowest was Piemontese (3.6), indicating that fat deposition occurs in different ways in different breeds (Robelin, 1986, Micol *et al.*, 1993).

Biometric measurements on the carcass also showed significant differences among breeds, especially for limb length, which varied from 64.4 cm in Highland to 86.3 cm in Holstein, and for internal depth of breast, which varied from 33.3 cm in Asturiana de los Valles to 43.3 cm in Holstein. There were also, significant differences in the limb index (F=30.9; p<0.001) and in the carcass blockiness index (F=96.7; p<0.001). The loin area was greatest for Asturiana de los Valles breed (52.7 cm<sup>2</sup>), followed by Piemontese (52.2 cm<sup>2</sup>), while Holstein presented the lowest value (35.0 cm<sup>2</sup>). These differences in the loin area were mainly due to differences in the dorso-ventral diameter, rather than in the medium-lateral dimension. Data for British breeds were not available.

From the dissection of the  $6^{th}$  rib it can be seen that different breeds show important differences in muscle (ranged between 58.1% in Holstein and 79.9% in Piemontese breeds) and fat percentages (ranged between 3.2% in Piemontese and 23.0% in Angus animals). These findings are in accordance with the carcass



measurements and conformation scores. The differences found in bone plus others were significant, but lower (F= 18.5) than those for fat and muscle (F= 52.4 and 56.9 respectively) thus, bone plus others ranged between 15.0% in Limousin and 23.1% in Holstein breeds. These results agree with the more precocious development of the bone tissue and of the "other" components, and subsequent their higher bio-stability, compared to fat and muscle tissues.

## Conclusions

We can conclude that breed is an important factor in determining carcass quality. The information that is being generated by this project will be important for devising breeding strategies to meet the demands of the market.

#### Acknowledgements

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fix SOD HOLS RED SIMM AV-NI JER AA HIG ASV ASM ΡI \* \* \* \* 13 \* 3 30 31 30 31 n as 189.81 347.24 350.74 248.64 317.14 322.15 345.00 348.74 244.69 324.70 371.45 for breed 25 11 20 Carcass weight (kg) (29.22)(19.70)(34.43)(22.94)(49.16)(30.14)(18.26) (11.55)(14.16)(15.80)(11.55)46.59 57.62 57.31 55.31 55.54 55.787 57.36 63.06 63.57 57.50 59.54 25 20 Dressing percentage (1.29) (1.89)(2.03)(2.31)(1.83)(1.07)(3.21)(3.02)(2.62)(1.7)(1.55)۲L, 7.67 23 7.64 8 2.29 9.25 2 5.52 3.57 2.15 2.32 3.85 3.24 3.86 Means, standard deviation and Kidney fat weight (kg) (2.27)(3.21)(0.63)(2.42)(1.77)(0.72)(0.40)(1.47)(1.28)(1.18)(1.79)10.53 4.36 22 10.43 7 8.33 3 4.92 5.00 7 7.33 12.10 7.58 8.00 11.48 Conformation score (1-18) (1.26)(0.58)(0.76)(0.58)(1.15)(2.80)(1.03)(1.23)(1.31)(1.40)(2.00)11.71 4.55 22 7.29 7 6.67 3 8.23 8.43 7 8.00 4.07 4.94 5.90 5.77 17 Fatness score (1-15) (1.15)(1.10)(2.69)(.83) (1.13)(0.00)(1.53)(1.16)(1.17)(0.57)(1.96)123.14 132.73 131.44 120.03 131.00 136.12 133.00 127.13 122.35 133.07 132.42 24 13 Carcass length (cm) (3.91) (6.56) (4.20)(7.50)(6.45)(4.87) (3.42)(16.67)(3.51)(5.93)(4.49)39.27 35.94 41.09 37.42 43.31 42.57 7 43.00 33.35 33.81 37.03 34.68 Internal depth of breast (cm) 13 (1.79)(0.93)(2.00)(1.82)(1.91)(1.92)(2.04)(1.47)(2.90)(1.57)(2.37)67.27 70.64 69.79 64.38 86.27 81.86 7 82.67 81.02 75.31 83.35 82.11 Limb length (cm) (2.31)(3.72)(2.52)(3.27)(2.79)(3.23)(2.99)(1.36)(2.52)(2.28)(2.30)cattle breeds. 25.93 28.00 30.22 25.73 27.77 31.05 26.54 Limb thickness (cm) ----(1.03)(0.67)(0.87)(2.62)(2.43)(2.38)(1.19)1.56 24 2.62 11 2.68 2.11 13 2.36 2.42 2.60 2.74 2.00 2.43 2.80 Blockiness index (kg/cm) (0.16)(0.10)(0.22)(0.15)(0.17)(0.10)(0.18)(0.24)(0.17)(0.27)(0.19)0.32 0.31 0.34 0.37 0.34 0.33 0.38 Limb index (cm/cm) ----(0.01)(0.02)(0.02)(0.03)(0.03)(0.03)(0.02)European Maximum diameter of the LT 9.51 9.997 9.23 9.77 9.31 8.57 8.99 --at 6<sup>th</sup> rib level (cm) (0.76)(0.80)(0.40)(0.91)(0.59)(1.15)(0.79)4.53 7 Minimum diameter of the LT 4.46 4.80 6.73 5.19 5.31 6.19 ---15 muscle at  $6^{th}$  rib level (cm) (0.46)(0.48)(0.52)(0.81)(0.65)(0.85)(0.66)quality traits in 35.03 36.58 38.67 52.71 35.85 40.31 48.68 LT area at the  $6^{th}$  rib level (cm<sup>2</sup>) -\_ \_ -(7.66)(6.65)(5.41)(5.31)(2.07)(8.25)(5.78)22.98 14.84 2 16.304 17.923 17.06 18.45 3 14.75 12.63 9.67 7.77 16 Fat percentage (6<sup>th</sup> rib dissection) (1.02)(6.35)(2.13)(4.22)(2.72)(4.56)(3.67)(2.43)(2.83)(4.26)60.40 58.73 <sup>3</sup> Muscle percentage (6<sup>th</sup> rib  $67.09^{2}$ 66.80<sup>4</sup> 64.22<sup>3</sup> 58.13 69.05 72.87 75.04 66.31 (1.93) (1.53)(3.69)(3.43)(5.74)(3.30)(2.59)(3.24)dissection) (3.95)(3.08)Carcass 16.62  $18.07^{2}$ 22.02 3 16.90 Bone and others percentage (6<sup>th</sup> 17.50 3 23.06 17.19 18.94 18.32 17.46 16 rib dissection) (0.51)(3.31)(0.39)(1.92)(2.02)(2.61)(2.15)(2.47)(1.74)(2.67)\* n varies in function of the considered variable because technical causes. Superscript indicates the n in each case.

MARC

28

307.52

(23.24)

58.75

(1.84)

1.36

(0.49)

11.11

(1.31)

4.96

(0.96)

123.87

(3.50)

39.61

(3.53)

72.79

(2.36)

31.13

(1.06)

2.48

(0.14)

0.43

(0.02)

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(0.84)

47.83

(5.35)

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(1.90)

70.07

(2.42)

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LIM

31

360.68

(20.85)

64.61

(0.98)

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(1.53)

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(1.16)

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(0.56)

126.66

(2.74)

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(1.82)

81.56

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(0.79)

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(0.14)

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(0.01)

9.63

(0.59)

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(4.12)

13.20

(2.33)

71.82

(2.87)

14.99

(1.58)

CHAR

30

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(27.42)

61.90

(1.65)

5.84

(1.24)

9.83

(1.23)

8.87

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133.02

(2.94)

34.43

(1.62)

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(1.87)

31.81

(1.12)

2.91

(0.18)

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(0.01)

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(0.88)

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(0.78)

43.91

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67.69

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16.88

(1.89)

F

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77.62

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12.43

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44.76

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84.75

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\*\*\*

18.52

\* \*\*

(p)

PIE

30

335.86

(28.59)

63.67

(1.81)

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(0.28)

14.57

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(0.86)

52.17

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(1.09)

79.91

(2.12)

16.86

(1.90)

JER.- Jersey; SOD.- South Devon; AA. - Aberdeen Angus; HIG. - Highland; HOLS. - Holstein; RED. - Danish Red Cattle; SIMM. -Simmental; ASV. - Asturiana de los Valles; ASM. - Asturiana de la Monta a; MV- Avile -Negra Ib rica; PHtenaica; PIE. -Piemontese; MARC.- Marchigiana; LIM.- Limousin; CHAR.- Charolais.

Table

ICoMST 2004 50<sup>th</sup> International Congress of Meat Science and Technology, Helsinki, Finland



## EUROPEAN CONSUMER ACCEPTABILITY OF LAMB MEAT FROM DIFFERENT ORIGINS AND PRODUCTION SYSTEMS

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#### Background

The international market for lamb meat tends to increase the commercial exchanges between geographical regions and countries. Europe is the greatest importer of lamb meat (Sañudo *et al.*, 1998a) with almost 50% of the world market. In this sense, South American countries are looking for new markets for exporting their red meat products. These products must compete with those produced locally. At the same time, each country or region in Europe produces a specific type of lamb according to its production system characteristics (breeds, environments, infrastructures, feeding resources, etc.). The particular characteristics of these systems determine the type of product that is commercialised in each region, according to liking, preferences and cooking habits of the consumers (Hernando *et al.*, 1996). Mediterranean countries, in general, produce lamb with lower carcass weights, while Central-Northern European countries produce animals with higher carcass weights. All these products are clearly appreciated in their own regions and producing to Jeremiah (1988), there are some reasons to believe that significant differences may exist in the palatability attributes of lamb from different geographical sources due to differences in genetics, nutrition, slaughter weight and age and the extent of post-mortem ageing.

## Objectives

The main objective of this study was to evaluate Spanish, German and British consumers' acceptability of lamb meat sourced from Uruguay and compared with their locally produced lamb at two different ageing periods.

#### Materials and methods

#### **Consumers**

Consumer tests were conducted in Spain, Germany and United Kingdom. In each of these three countries, two hundred consumers (in two different places of 100 consumers each) evaluated 4 different lamb meat samples: 2 local and 2 from Uruguay. Overall consumers were stratified in terms of sex, age and education level. Gender classification was almost equal (male-51%, female-49%), with smaller differences within each country. On the other hand, consumers were mostly concentrated on the two medium ranges of age (26-40 and 41-60 years of age, 32% and 39 % respectively), with an important variation in the extreme groups in each country; in average 17% (18-25 years of age) and 12% (61-75 years of age). The education level of the sampled population was in general very high (56% of people with University or college education level), 26% was located at the school to age 16 and 18% was at the school to age 18.

#### Animals and meat samples

Uruguayan lambs comprised light and heavy Corriedale lambs  $(11.1\pm1.4 \text{ kg} \text{ and } 19.6\pm2.2 \text{ kg} \text{ carcass weight},$  respectively) reared on a pure extensive improved sward system and the meat aged for 20 days. In the case of the European lambs only one type by country was used, representative of each country where the comparative consumer tests were performed, aged for either 7 or 20 days. Samples taken from United Kingdom represented a common commercial lamb type  $(22.8\pm1.7 \text{ kg} \text{ carcass weight})$  reared on grass-based



system with strategic use of concentrate. German samples were taken from crossbreed between Suffolk or Schwarzköpfe x Merino Landschaf ( $23.2\pm3.65$  kg carcass weight) reared on grass complemented with concentrates. In Spain, lambs came from the Rasa Aragonesa breed, produced under an intensive system using concentrates and cereal straw *ad libitum* ( $10.2\pm0.6$  kg carcass weight).

All meat samples were taken from the loin (*M. longissimus lumborum*). After thawing, they were sliced into 2 cm-thick steaks and grilled until the internal temperature reached 72°C. Consumers evaluated tenderness, flavour and overall acceptability on 8-points category scales.

#### **Statistics**

Consumer test results were separately analysed for each country. The analysis of variance was performed using the GLM procedure of SAS for Windows version 8.1 (SAS, 2000). Lamb type was included as a fixed effect and consumer as a random effect. The session effect had been previously corrected.

## **Results and discussion**

Tables 1, 2 and 3 show the results of the consumer tests carried out in Spain, United Kingdom and Germany, respectively.

Table 1. - Tenderness, flavour and overall acceptability of lamb by Spanish consumers. Least Square Means and Root MSE.

Lamb type	Tenderness*	Flavour*	Overall acceptability*
Light Uruguay	6.1	5.7 ab	5.8
Heavy Uruguay	6.2	5.5 bc	5.6
Spain 7d ageing	6.3	5.8 a	5.8
Spain 20d ageing	6.3	5.4 c	5.6
RMSE	1.28	1.56	1.44

a, b, c: LSMeans with different letters within tenderness, flavour and overall acceptability are statistically different (P<0.05). \*: from 1, very tough and extremely disliked flavour and acceptability, to 8, very tender and extremely liked flavour and acceptability.

In Table 1, it can be observed that Spanish consumers preferred the flavour of the Spanish lamb meat aged for 7 days and of the Uruguayan light lamb (P<0.05), without differences in tenderness and overall acceptability (P>0.05). This could partially be explained by the similarity of live weight and age between the Spanish and the Uruguayan light lamb, even though they came from very different production systems.

German consumers (Table 2) gave higher ratings of tenderness to the Uruguayan heavy lamb and the German lamb aged for 20 days, with higher flavour ratings to both German lamb samples, and the highest overall acceptability given to the German lamb type aged for 20 days (P<0.05). At the same time, the flavour and overall acceptability of Uruguayan light lambs were the least preferred (P<0.05). In general, it seems clear that German consumers preferred their own products and those that have some similar characteristics to them (Uruguayan heavy lamb), even being consumers without or with a very small experience in lamb meat consumption. They rejected meat from Uruguayan light lamb, which was the only unweaned type. Thus, Kemp *et al.* (1981) found that differences in flavour between two groups of lambs could be explained by differences in feeding regimens when comparing lambs fed exclusively with maternal milk versus lambs supplemented with commercial pellets and hay *ad libitum*.



Lamb type	Tenderness*	Flavour*	Overall Acceptability*
Light Uruguay	6.1 b	5.7 c	5.7 c
Heavy Uruguay	6.6 a	6.0 b	6.1 b
German 7d ageing	6.2 b	6.1 ab	6.1 b
German 20d ageing	6.7 a	6.3 a	6.4 a
RMSE	1.14	1.18	1.07

 Table 2. - Tenderness, flavour and overall acceptability of lamb by German consumers. Least Square Means and Root MSE.

a, b, c: LSMeans with different letters within tenderness, flavour and overall acceptability are statistically different (P<0.05). \*: from 1, very tough and extremely disliked flavour and acceptability, to 8, very tender and extremely liked flavour and acceptability.

For the UK consumers (Table 3), flavour and overall acceptability of the British lamb aged for either 7 or 20 days and of the Uruguayan heavy lamb were similar, and they obtained the higher ratings(P<0.05). The highest tenderness ratings were for the Uruguayan heavy lamb and British lamb aged for 7 or 20 days (P<0.05), while the lowest ratings were given to the Uruguayan light lamb (P<0.05). In general it can be suggested that British consumers were able to recognise their own products (grass fed) for lamb meat flavour and overall acceptability. These results were probably due to the strong influence that age (older animals) and production and feeding systems has on flavour, and the relationship between flavour and consumer acceptability. In the same way, Sañudo *et al.* (1989), working with lambs of different origins (Spanish light lamb, New Zealand and French light lambs) tested in Spain, found that New Zealand lamb meat obtained the highest values for flavour intensity and the lowest ratings for overall acceptability. Crouse *et al.* (1983) and Solomon (1980) reported similar results.

Table 3. - Tenderness, flavour and overall acceptability of lamb by British consumers. Least Square Means and Root MSE.

Lamb type	Tenderness*	Flavour*	Overall Acceptability*
Light Uruguay	5.6 c	5.4 b	5.3 b
Heavy Uruguay	6.9 a	6.1 a	6.3 a
UK 7d ageing	6.5 b	6.1 a	6.2 a
UK 20d ageing	6.7 ab	6.2 a	6.3 a
RMSE	1.28	1.42	1.35

a, b, c: LSMeans with different letters within tenderness, flavour and overall acceptability are statistically different (P<0.05). \*: from 1, very tough and extremely disliked flavour and acceptability, to 8, very tender and extremely liked flavour and acceptability.

In general, these findings are probably related to the consumption habits of the different consumers evaluated, which determine a lower acceptance of unfamiliar products. This can be clearly seen when Uruguayan light lamb was compared to German and British lamb types or when Uruguayan heavy lamb was compared to Spanish lamb samples. In a previous report using British and Spanish lamb carcasses, Sañudo *et al.* (1998b) showed that both British and Spanish panels found the odour and flavour intensity higher in the British carcasses and juiciness higher in the Spanish lamb types. However, the panels differed in their ratings of flavour quality and overall appraisal. The British panel preferred the flavour and overall liking of British lamb, whereas the Spanish panel preferred the flavour and overall appreciation of Spanish lamb.



## Conclusions

Lamb meat acceptability depends on the consumption habits of the consumers, at least in countries with relatively high lamb consumption such as Spain and United Kingdom. Also, lamb meat tenderness was improved by ageing period, although only significantly in Germany, and, in general, more tender lamb meat was associated with higher acceptability at longer ageing times (20 days), especially in the older and heavier lamb types.

Overall acceptability seems to be better related to flavour than tenderness scoring, and this is in agreement with previous European studies (Dransfield *et al.*, 1984) that showed increased focus on flavour and overall acceptability when all the meat being compared was tender.

#### Acknowledgements

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## THE RELATIONSHIP BETWEEN CIE L\* AND PH AT 1 DAY POSTMORTEM IN PORCINE SEMIMEMBRANOSUS MUSCLES HARVESTED FROM NATIONAL PORK DEVELOPMENT HOGS

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#### Background

Pork quality is defined by CIE L\* (lightness), ultimate muscle pH, and drip loss in Kauffmann et al. (1992). Pale, Soft, and Exudative (PSE) pork is a quality classification characterized as being very light colored, soft, and watery. Such meat is classified as low quality and undesirable to consumers due to its poor appearance, texture, and palatability (Pearson and Gillett, 1996). PSE meat also exhibits poor water holding capacity and texture in processed products (Solomon et al. 1998). PSE meat originates from a rapid decline in pH postmortem (45 min) at high muscle temperature (Briskey and Wismer-Pedersen, 1961). PSE meat was identified (Kauffmann et al, 1992) by ultimate pH< 5.6, CIE L\* Value > 50, and drip loss > 5%, and red, firm, and non-exudative (RFN) or normal pork was classified as ultimate pH=5.6-5.9, 42<CIE L\*< 50, and drip loss < 5 %. It is not guaranteed that a sample with a CIE L\* value > 50 is PSE. However, in theory, the above-listed classifications would allow for two possible relationships between CIE L\* value and ultimate pH. There would be either a strong linear relationship between CIE L\* value and pH over the whole range of values or samples with a pH greater than 5.6 would generally have a CIE L\* value greater than 50 and samples with a pH less than 5.6 would have a CIE L\* value less than 50. For either example, it may be possible to utilize ultimate pH to predict with accuracy if a porcine semimembranosus muscle CIE L\* value is greater than or less than 50. Unpublished data from the industry indicates that there may not be a clear relationship between ultimate pH and CIE L\* in pork. This research was performed to first test the hypothesis that there is a relationship between CIE L\* and ultimate pH for national pork development hogs and then to characterize that relationship if it exists.

## Objectives

The main objective of this research is to demonstrate if there is a relationship between ultimate pH and lightness of porcine *semimembranosus* muscles in National Pork Development Hogs. A second objective is to utilize logistic regression to predict if the CIE L\* value is greater or less than 50 based on ultimate pH.

## **Materials and Methods**

Porcine *semimembranosus* and *adductor* muscles (n=384) were obtained from a pork processing plant in Virginia (United States) on 15 separate occasions to obtain a good representation of sample color. Samples were selected based on visual color as an attempt to obtain half of the samples as pale and half as normal. All samples were taken from National Pork Development (NPD) pork carcasses produced from market age pigs that weighed 110-125 kg. CIE L\* values were measured using a chroma meter (Model CR-200, Minolta Camera Co., Ltd., Osaka, Japan) at three similar anatomical locations on each muscle. The chroma meter was calibrated using a standard Minolta calibration plate (white plate, No. 20933026; CIE L\* 97.91, a\* -0.70, b\* +2.44) each time prior to testing. The pH of each *semimembranosus/adductor* muscle was also taken in triplicate by removing three 2-g samples from the three same similar anatomical locations that color was measured and homogenized (Virtishear Model.225318, The Virtis Company, Inc., Gardener, NY) for 10-20 s (3 short bursts) in 20 ml of distilled deionized water. pH was measured for the individual samples with a calibrated pH meter (Model AR25, Fisher Scientific, Pittsburgh, PA).

## Statistical Analysis

The correlation coefficient was determined for the relationship between CIE L\* and pH for all samples, those with CIE L\* values less than 50, and CIE L\* values greater than 50 (SAS 8.2, 2001). Logistic regression



was utilized to determine how well ultimate pH would predict if a sample had a CIE  $L^* > 50$  or a CIE  $L^*$  value <50 (SAS 8.2, 2001).

#### **Results and Discussion**

There is a strong negative correlation (r= - 0.83) between ultimate pH and CIE L\* values for porcine semimembranosus muscles (Figure 1). This relationship is accurate, but is also misleading. Separation of the data into two categories including ultimate pH>5.6 and ultimate pH<5.6, established a correlation of - 0.45 and -0.45, respectively (Figures 2 and 3). This drastic decrease reveals that the relationship between CIE L\* and ultimate pH is misleading. Furthermore, through the removal of outliers from the data, the correlation decreases from -0.45 to -0.40 for pH below 5.6 and from -0.45 to -0.046 for samples above a pH of 5.6. These results demonstrate that there is not a linear relationship between the data and that logistic regression may be appropriate to determine the relationship between a samples ultimate pH and CIE L\* value.

The logistic regression analysis demonstrated that ultimate pH is effective (p < 0.05) in determining whether the CIE L\* value will be either greater or less than 50. The logistic regression model predicted that if the pH was less than 5.6, there is a 0.962 probability that the CIE L\* would be above 50. At a pH above 5.6, the predicted probability that the CIE L\* value is below 50 was 0.905. This research reveals that ultimate pH is a good predictor for whether CIE L\* will be above or below 50. The relationship between these two variables is that of a categorical distribution where the probability that a sample would have a CIE L\* value greater or less than 50 can be predicted based on pH value. Furthermore, predictions of whether CIE L \* is greater or less than 50 can be predicted at any pH (Figure 4). Predictions can actually be made for any set of categories that are desired. Figure 4 reveals that as the ultimate pH approaches 5.4 and 5.9, the probability that a sample has a CIE L \* value less or greater than 50 approaches 1. Therefore, from 5.9-7.0, the probability that the CIE L\* value will be below 50 is 1, and the predicted probability that any sample with a pH below 5.4 having a CIE L\* value is 1. At pH values of 5.6 and 5.7, the probabilities that a sample will be above 50 in CIE L\* is 0.81 and 0.39, respectively (Figure 4). This evaluation reveals that there is a higher probability that a sample is pale than normal in this pH range. Therefore, ultimate pH is a useful indicator of product paleness when pH is higher than 5.8 and less than 5.5, but is ineffective in classifying porcine semimembranosus color from NPD hogs when the pH is between 5.5 and 5.8.

#### Conclusions

The ultimate pH of porcine *semimembranosus* muscles correlates well with CIE L\* values. CIE L\* values can be explained by ultimate pH when CIE L\* values are divided into a categorical variable. Logistic regression is a useful tool in explaining the relationship between ultimate pH and CIE L\* values.

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Figure 1: The Relationship Between ultimate pH and CIE L\* value of porcine semimembranosus muscles (n=384) of National Pork Development (NPD) hogs.



Figure 2: The relationship between ultimate pH and CIE L\* for semimembranosus muscles (pH<5.6, n=184) from National Prk Development Hogs





Figure 3: The relationship between ultimate pH and CIE L\* for semimembranosus muscles (pH>5.6, n=200) from National Pork Development Hogs



Figure 4: The Predicted probability that a porcine *semimembranosus* muscle from a National Pork Development (NPD) hog with a certain ultimate pH will have a CIE L\* less than or greater than 50.



## THE EFFECTS OF RUMINALLY-PROTECTED DIETARY LIPID ON THE FATTY ACID COMPOSITION AND QUALITY OF BEEF MUSCLE

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## Background

Improving the nutritional value of beef by reducing saturated fat and increasing the content of polyunsaturated fatty acids (PUFA) is an important research target. Rumen biohydrogenation of dietary PUFA limits the ability to manipulate the ratio of polyunsaturated:saturated fatty acids (P:S), which for beef is typically 0.06 - 0.1 (Wood *et al.*, 2003). However, feeding a ruminally-protected, PUFA-rich, lipid supplement produced from soya beans, linseed and sunflower oils mixed to give a 2.4:1 ratio of 18:2*n*-6:18:3*n*-3 resulted in major improvements in the P:S ratio (Scollan *et al.*, 2003) with only small associated changes in the colour shelf life and sensory attributes of the meat (Enser *et al.*, 2001). However, in that study, the protected lipid supplement (PLS, with an 18:2*n*-6:18:3*n*-3 ratio of 2:4:1) was less effective in increasing the deposition of *n*-3 relative to *n*-6 PUFA, resulting in a less favourable *n*-6:*n*-3 ratio in beef longissimus muscle. It is hypothesised that the ratio of *n*-6:*n*-3 PUFA in the lipid supplement has a large impact on the deposition of *n*-6 relative to *n*-3 PUFA in muscle.

## Objectives

To determine the effects of including a runnially protected lipid supplement, in the diet of beef cattle, with an 18:2n-6:18:3n-3 ratio of 1:1 on the fatty acid composition of muscle neutral and phospholipids and to relate this to meat shelf-life (colour and lipid oxidation) and flavour.

#### Materials and methods

Thirty two Charolais steers (initial live weight 507 (s.e. 10.3) kg) were fed on *ad libitum* grass silage plus one of four concentrates in which the lipid source was either Megalac (Mega, rich in palmitic acid; 16:0) or PLS (soya beans, linseed and sunflower oils resulting in a 1:1 ratio of 18:2n-6:18:3n-3): Concentrate 1, (Mega, control) contained 139g/kg Mega; Concentrate 2, (PLS1) contained 67g/kg Mega with 400 g/d PLS fed separately; Concentrate 3, (PLS2) contained 24g/kg Mega with 800 g/d PLS fed separately, Concentrate 4, (PLS3) contained no Mega and 1000 g/d PLS fed separately. Supra-nutritional levels of vitamin E were included in all diets (350 IU/kg concentrate). At 48h post-mortem, samples of m. longissimus thoracis at the 11th rib were removed and blast frozen for fatty acid analysis. Other samples of m. longissimus lumborum were removed and conditioned for 10 days in vacuum packs at 1°C, then a joint was frozen for subsequent organoleptic assessment and steaks cut and packed in a modified atmosphere (O<sub>2</sub>:CO<sub>2</sub>, 75:25). These were displayed for 10 days at 4°C under 700 lux for 16h out of each 24h to simulate commercial retail display. Colour was measured daily using a Minolta Chroma Meter. Lipid oxidation was determined as thiobarbituric acid reacting substances (TBARS) (Tarladgis et al., 1960) after 10d display. After thawing, steaks were cut from the frozen joint, grilled to an internal temperature of 74°C and sensory assessments made by a 10 member trained taste panel using 100mm unstructured line scales. Lipid was extracted using chloroform/methanol and the neutral and polar lipids separated by silicic acid column chromatography. Fatty acid methyl esters were prepared by alkaline hydrolysis followed by methylation with diazomethane and analysed on a CP Sil 88, 100m x 0.25mm ID column (Chrompack, UK).

## **Results and discussion**

Total lipid, total neutral lipid and total phospholipid fatty acids were not influenced by diet (Tables 1, 2 and 3). The content of the main saturated fatty acids (14:0, 16:0, 18:0) were not different in either lipid fraction, but 18:1*n*-9 was reduced in the neutral lipid (Table 1). On average, feeding PLS increased the content of



18:2*n*-6 and 18:3*n*-3 by a factor of 3.2 and 5.9, respectively in the neutral lipid. In phospholipid, on average, PLS increased the content of 18:2*n*-6 and 18:3*n*-3 by 1.86 and 2.31, respectively. Interestingly, in contrast to the neutral lipid, no further increases in either 18:2*n*-6 or 18:3*n*-3 were noted in phospholipid after the first increment of PLS (Table 2). These increases in C18 PUFA in phospholipid were associated with reductions in oleic acid, 18:1*n*-9 and DPA, 22:5*n*-3. The percentage of 18:2*n*-6 and 18:3*n*-3 in muscle total lipid on the highest level, PLS3, was 3.2 and 5.9%. This compares with 9.3 and 1.9% for 18:2*n*-6 and 18:3*n*-3 at the highest inclusion of a PLS (with a 2.4:1 18:2*n*-6 and 18:3*n*-3) in the study by Scollan *et al.* (2003). The P:S ratio increased markedly (P < 0.001) while the *n*-6:*n*-3 ratio was decreased (P < 0.001; Table 3).

The increase in PUFA in the meat, with increasing PLS fed, was associated with increased susceptibility to lipid oxidation as reflected in higher TBARS (Table 3) and colour deterioration (Figure 1). Colour acceptability (saturation index > 18) was decreased by approximately 2 days for PLS3 relative to Mega. The high levels of vitamin E fed to counteract the effects of increasing PUFA in the tissue were not sufficient to negate the negative effects on colour shelf life or lipid oxidation. There was a trend for the tenderness of meat samples to increase with an increase in the amount of protected lipid fed (Table 4). PLS2 and PLS3 samples were significantly juicier than samples from PLS1. There was a trend for beefy flavours to decline with increased supplementation. However, abnormal flavour increased significantly with increasing PLS. This was associated with a trend for greasy to increase, a significant rise in rancid and a trend for vegetable/grassy and dairy notes to increase. These trends in abnormal flavours resulted in a significant reduction in overall liking for PLS2 and PLS3 compared to PLS1 and control.

The fatty acid changes had marked effects on lipid oxidation, colour shelf life and beef flavour. These results provide very clear evidence that these characteristics are inter-related. Increasing concentrations of PUFA in meat to the extent seen here would therefore not be a commercial proposition without further antioxidant protection. This would include careful meat processing procedures as anything that increases oxidative susceptibility such as mincing or cooking would further increase lipid oxidation.

## Conclusions

The protected lipid supplement rich in 18:2n-6 and 18:3n-3 resulted in large increases in these fatty acids in beef muscle. This resulted in a favourable increase in the P:S ratio and a beneficially lower n-6:n-3 ratio than that observed in our previous study (Scollan *et al.*, 2003). The changes in fatty acid content of the meat had marked effects on lipid oxidation, colour shelf life and beef flavour. The high levels of vitamin E fed were insufficient to significantly reduce the negative effects of increasing PUFA in the meat on lipid stability and colour shelf life. There is a need to determine the role of antioxidants in enhancing the stability of meat which has enriched levels of PUFA.

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	Mega	PLS1	PLS2	PLS3	s.e.d.	Р
14:0 myristic	123.9	121.6	119.2	114.0	23.70	NS
16:0 palmitic	1216	1215	1155	1097	199.7	NS
18:0 stearic	661	666	712	697	112.0	NS
18:1 <i>trans</i>	102	106	109	128	18.1	NS
18:1n-9 oleic	1521	1595	1559	1563	269.0	NS
18:1 cis vaccenic	56	58	58	58	9.6	NS
18:2 n-6 linoleic	42 <sup>a</sup>	109 <sup>b</sup>	135 <sup>b</sup>	158 <sup>b</sup>	18.7	0.001
18:3 n-3 α-linolenic	15 <sup>a</sup>	69 <sup>b</sup>	88 <sup>bc</sup>	110 <sup>c</sup>	12.5	0.001
CLA cis-9, trans-11 C18:2	18.7	22.8	23.0	29.8	4.98	NS
Total fatty acids	4137	4370	4352	4370	715	NS

Table 1. Fatty acid content (mg/100g muscle) of neutral lipid fraction of *M. longissimus thoracis* 

Table 2. Fatty acid content (mg/100g muscle) of phospholipid fraction of M. longissimus thoracis

	Mega	PLS1	PLS2	PLS3	s.e.d.	Р
14:0 myristic	2.36	3.57	2.52	2.52	0.971	NS
16:0 palmitic	89	97	83	78	8.7	NS
18:0 stearic	56.4	64.8	60.8	61.5	4.51	NS
18:1 <i>trans</i>	3.7	4.6	4.2	4.4	0.55	NS
18:1n-9 oleic	123 <sup>c</sup>	74 <sup>b</sup>	51 <sup>ab</sup>	49 <sup>a</sup>	12.0	0.001
18:1 cis vaccenic	11.9 <sup>b</sup>	11.0 <sup>b</sup>	8.2 <sup>a</sup>	8.6 <sup>a</sup>	1.02	0.002
18:2 n-6 linoleic	78.9 <sup>a</sup>	146.7 <sup>b</sup>	144.4 <sup>b</sup>	146.9 <sup>b</sup>	9.46	0.001
18:3 n-3 α-linolenic	13.1 <sup>a</sup>	32.7 <sup>b</sup>	29.6 <sup>b</sup>	28.5 <sup>b</sup>	2.22	0.001
CLA cis-9, trans-11 C18:2	1.23	1.35	1.16	1.17	0.187	NS
20:4 n-6 arachidonic	26.9	27.5	24.5	24.5	1.97	NS
20:5 n-3 eicosapentaenoic (EPA)	12.4	14.8	13.5	14.8	1.10	NS
22:5 n-3 docosapentaenoic (DPA)	20.9 <sup>b</sup>	20.9 <sup>b</sup>	17.4 <sup>a</sup>	16.5 <sup>a</sup>	1.49	0.009
22:6 n-3 docosahexaenoic (DHA)	1.89	1.81	1.53	1.56	0.272	NS
Total fatty acids	548	606	528	525	42.0	NS

Table 3. Total fatty acids (mg/100g muscle), nutritional indices of total lipid of *M*.longissimus thoracis and thiobarbituric acid ((TBARS) mg malonaldehyde/Kg meat) values for *M*.longissimus lumborum after 10 days simulated retail display in modified atmosphere packs

	Mega	PLS1	PLS2	PLS3	s.e.d.	Р
Total fatty acids <i>n</i> -6: <i>n</i> -3 ratio P:S ratio	4685 2.27 <sup>c</sup> 0.07 <sup>a</sup>	4976 2.02 <sup>b</sup> 0.177 <sup>b</sup>	4880 2.00 <sup>b</sup> 0.199 <sup>c</sup>	$4895 \\ 1.88^{a} \\ 0.218^{d}$	737 0.055 0.0179	NS 0.001 0.001
TBARS Day 10	0.54 <sup>a</sup>	2.04 <sup>b</sup>	4.17 <sup>c</sup>	4.03 <sup>c</sup>	0.665	0.001





**Figure 1.** The effect of days displayed upon the change in colour saturation (±stdev) of modified atmosphere packed loin steaks, from animals fed varying levels of protected lipid supplement.

<b>Table 4.</b> Effect of Protected Lipid Supplement on sensory values of grilled loin steaks
Values are the means derived from analysis of variance with Supplement and Assessor as factors; panels are
treated as a 'block structure' with 8 replications.

	Mega	PLS1	PLS2	PLS3	s.e.d.	Р
Toughness	41.8	43.4	44.2	38.7	2.51	NS
Juiciness	36.5 <sup>ab</sup>	40.3 <sup>b</sup>	33.5 <sup>a</sup>	35.0 <sup>a</sup>	2.29	0.05
Beef	25.1	25.2	23.0	21.0	1.79	NS
Abnormal	18.5 <sup>a</sup>	17.9 <sup>a</sup>	$20.9^{ab}$	24.4 <sup>b</sup>	2.10	0.01
Greasy	15.3	14.4	16.5	16.2	1.78	NS
Bloody	4.7 <sup>a</sup>	8.6 <sup>b</sup>	6.4 <sup>ab</sup>	4.7 <sup>a</sup>	1.55	0.05
Livery	7.2	5.2	6.1	5.0	1.82	NS
Metallic	4.9	7.4	7.8	5.3	1.26	NS
Bitter	3.9	2.3	3.0	2.5	1.25	NS
Sweet	1.9	3.2	2.5	1.5	0.77	NS
Rancid	1.4 <sup>a</sup>	1.4 <sup>a</sup>	$2.3^{ab}$	3.8 <sup>b</sup>	0.76	0.01
Fishy <sup>†</sup>	1.5	1.3	2.2	4.2	1.34	NS
Acidic	10.8	10.8	10.3	9.0	2.02	NS
Cardboard	3.9	5.0	5.3	3.3	1.44	NS
Vegetable/grassy	7.5	8.6	8.6	10.5	1.90	NS
Dairy	8.0	8.6	5.5	11.1	2.11	NS
Hedonic						
Overall liking	23.2 <sup>c</sup>	22.8 <sup>bc</sup>	19.2 <sup>a</sup>	19.6 <sup>ab</sup>	1.69	0.05

Figures with the same superscript do not differ significantly

<sup>†</sup>Interaction between type and assessor, therefore vr, sed and sig are recalculated.



## IMAGE ANALYSIS FOR CHARACTERIZATION OF THE INTRAMUSCULAR CONNECTIVE TISSUE IN MEAT

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#### Background

Image analysis is a promising approach to characterize the spatial organization of the intramuscular connective tissue (IMCT) using morphological features. A common approach is based on segmentation which consists in subdividing an image into its constituent parts (Pal and Pal, 1993). In meat science segmentation has been principally used to quantify fat distribution on steaks and to determine skeletal maturity of carcass (Tan, 2004). We developed and applied an adaptative segmentation for extracting IMCT thus permitting characterization of spatial organization from visible images of histological sections of muscles. It can also be used to quantify morphometrical parameters on segmented objects, such as size or network hierarchy parameters.

#### Objectives

This paper describes a practical approach, called Fuzzy Cards Thresholding (FCT), based on building reference maps: a) to set up thresholding rules based on the features calculated from the intensity histogram, b) to quantify the precision of the segmentation and to compare it with other methods. Once the optimal segmentation was obtained, morphometrical parameters characterizing the distribution of IMCT were quantified and compared within several muscle types.

#### Materials and methods

<u>Animals and sample preparation</u> Muscle samples were taken from 2-year-old Aubrac heifers (n=4). *Biceps femoris* (BF), *Infraspinatus* (IS), *Longissimus thoracis* (LT) and *Pectoralis profundus* (PP) were chosen for their different architectures, including fiber bundle size, IMCT thickness and hierarchy. Samples were aged 8 days *post mortem* at 4°C before cutting and staining. Muscles samples of  $20 \times 20 \times 10 \text{ mm}^3$  were frozen with isopentane chilled by liquid nitrogen at -160°C. They were cut to make transversal sections of 0.01 mm thickness. Perimysial tissue was revealed with red Sirius stain, specific for collagen, which is the main connective tissue component. A second group of animals (8 Charolais cows) was submitted to the same treatment and used for collagen amount determination and to study the differences between muscles.

<u>Optimization of thresholding using fuzzy reference maps</u> The process is based on 4 steps: a) to build the fuzzy reference maps, b) to threshold the map at different confidence levels, c) to define the optimal threshold which minimizes the difference between the thresholded image and the reference card, and d) from n cards obtained on different animals and muscles, to define the thresholding rules according to the histogram features of the images.

Each sample image was digitalized with a transmitting light box and a CCD camera JAI CV-M300 coupled with a macroscopic objective leading to a pixel size of  $6.8 \cdot 10^{-4}$  mm<sup>2</sup> (figure 1). Visilog software (Noesis, France) has been used for image processing. The grey level histogram, in the whole field of view, was computed and features such as the mean, standard deviation, skewness and kurtosis were calculated from the histogram moments. Ten test images of 50 mm<sup>2</sup> of section were displayed at random to a panel of 20 non-trained judges. The judges had to draw (with a one-pixel-thick and 0 grey level line) what they thought to be IMCT (figure 2b). The thickest segments which everyone could identify had already been labelled by thresholding at a high level. The work of the judges was to trace the thinnest elements of the network which are more difficult to identify. Their digital drawings were averaged for each image to obtain the fuzzy reference maps for the perimysial network (figure 2c).

By construction, a fuzzy reference map is in grey levels. It has to be binarized in order to be compared with the result of our adaptive threshold method. When the reference map is thresholded, the resultant binary image  $(S_{\epsilon})$  is composed of the pixels selected by a percentage of judges called confidence level,  $\epsilon$ . A high  $\epsilon$  ( $\epsilon$ )



> 50%) corresponds to a consensual set of IMCT, which favours points selected by most of the judges. In this case, S<sub> $\varepsilon$ </sub> represents the thick segments, which are distinguished without ambiguity (figure 2d). On the contrary, for  $\varepsilon < 50\%$ , the thresholded reference map includes the thinner segments (figure 2e).

To evaluate the quality of the segmentation for a given  $\varepsilon$ , the total error E has been computed as a performance index. It is the sum of two error terms, the fraction *Eoo* of pixels of the image belonging to the object (perimysium) which are falsely attributed to the background and the fraction *Eob* of pixels of the image belonging to the background, which are falsely attributed to the objects. As *Eob* decreases and *Eoo* increases as function of threshold, *E* exhibits a local minimum, which is the optimal value of t (t\*), that is a compromise between *Eoo* and *Eob* (figure 3). The t\* value is obtained by minimizing the total error E which is the fraction of pixels which have been segmented improperly and it is determined for an a priori fixed confidence level.

Using SAS software (Statistical Analysis Systems Institute, 1995), a stepwise multiple linear regression (GLM procedure) was performed between t\* values and histogram features for each reference image (n=10) at different confidence levels (from 30% to 80%). Thus models were obtained that could predict the thresholding level of all the images taken in the same conditions as our test images, for each confidence level.

<u>Validation of the method</u> The validation was based on a comparison of our method with a reference method already optimized and proposed by Rosin (2001) for the segmentation of this type of images showing a unimodal histogram (RM). It considered a dominant population in the image that produces one main peak relative to the secondary population. This latter class may or may not produce a discernible peak, but needs to be reasonably well separated from the bulk peak to avoid being swamped by it.

Eight sections from 4 muscles of Charolais cows were used. They were stained, digitalized and then the collagen amount was determined as the amount of stain fixed by the collagen fibers and then eluted. (Lopez De Leon and Rojkind, 1985).

## **Results and discussion**

<u>The reference maps</u> Each pixel of the reference map had a value corresponding to a grey level from 0 (black) to 255 (white). A black pixel (0) had a nil probability to belong to the perimysium (none of the judges designated it). A white pixel (255) had the maximal probability (all judges designated it). So the maps were fuzzy because a pixel was attributed a probability of belonging to the perimysium and not a cut decision. The decision happened when a confidence level was chosen. The pixels of the maps can only take a discrete number of intensities equal to the expert number. The maps give information on the location of the connective tissue and also on the error (confidence level,  $\varepsilon$ ) made by keeping this pixel in the "perimysium" class of objects. In this way, the maps give different patterns according to  $\varepsilon$ . We observed that there was a natural correspondence between  $\varepsilon$  and the thickness of the thresholded segments.

<u>Thresholding rules</u> For each image the optimal threshold was determined at different confidence levels as shown in table 1. The optimal threshold increased when  $\varepsilon$  decreased. It segmented lighter and thinner objects, because low  $\varepsilon$  values accept pixels with a lower belonging probability.

The results of the multiple linear regressions are shown in table 1. They are all linear combinations of the mean and standard deviation or skewness. Models are very informative for each  $\varepsilon$  value from 30% to 80%. At high confidence levels (70%, 75% and 80%) which correspond to the thick network, the standard error was superior to 3 grey levels. Indeed the segmentation quality was less sensitive to the threshold choice because of the high contrast between myofibers and IMCT. The effective error had fewer consequences than at a small  $\varepsilon$  values, because it was far from the over segmentation zone.

<u>Comparison with Rosin's method</u> The performance index decreased when the confidence level increased and the quality of segmentation improved (figures 4 and 5). Indeed for high  $\varepsilon$  values, there were fewer pixels to detect and a higher contrast. RM performance index varies more than with our method (29.6% and 22.2% respectively). This was due to the fact that our method (FCT) adapts the threshold to the confidence level, while RM is adaptative to image context and gives a unique threshold whatever the value of  $\varepsilon$ . RM *Eoo* (segmentation on thin segments), was almost 2 fold higher at low  $\varepsilon$ , which indicated that it did not take into account thin segments. With FCT, *Eoo* and *Eob* decreased when  $\varepsilon$  increased and the indexes were lower than for RM. A better segmentation was obtained with FCT which could be adapted to  $\varepsilon$ . Indeed when  $\varepsilon$  decreased, the thinner frame had to be segmented, so the threshold needed to go towards the lighter grey



levels. *Eob* is higher but the performance index stayed inferior to RM. In fact, *Eob* is inherent to the quality of the image, and in particular to its background homogeneity.

The segmentation performance did not actually seem to be largely improved by decreasing  $\varepsilon$ . However, a slight difference in the performance index (E) could imply a big difference in the segmentation. Only few pixels belonging to the thinnest connective network were detected, but they may provide valuable information on the hierarchy of the network (figure 5). However  $\varepsilon$  was limited to 55% for this experiment. Comparison with collagen amount determination

 $R^2$  was the highest at high confidence levels and it decreased markedly with  $\varepsilon$  (table 2). Thick networks seemed to have more influence on collagen content, since they represented a large number of pixels. Moreover, detection of parasite pixels increased because of the error of the thresholding rules, and it depreciated the correlation when  $\varepsilon$  decreased.

Discriminating parameters between muscles Three variables were calculated: the area of the segmented network, its length and its average thickness, considered as the ratio between the area and the length (table 3). For  $\varepsilon = 80\%$  (thickest segmentation), the thickness of the segments showed differences between muscles which present visually a thick network (BF, PP), as shown in figure 1, whereas there was no difference in thickness between muscles with thin connective tissue (IS, LT). In contrast, for  $\varepsilon = 55\%$  (segmentation of the whole network), IS and LT were discriminated. The values of thickness were in the same range as those published by Geesink et al (1995). Thickness seemed to be related with tenderness because BF and PP (tough muscles) have the thickest networks. The area of the network could not discriminate BF and PP, as the collagen amount did. BF had the largest area of IMCT at 80%, and the smallest at 55%. It had a thick network and few thin segments. In contrast, IS had the largest area of IMCT at 55%. The total length of the network, which was homogeneous to the network branching, segregated the 4 muscles at high confidence levels. PP and IS which showed the greatest degree of branching had longer networks than LT and BF.

## Conclusions

We have developed an automatic and validated approach for global thresholding of unimodal images of the intramuscular connective tissue network in muscle and meat. Our method gives objective variables for characterization of the IMCT, and gives further insights into those factors which determine the tenderness of meat by studying the IMCT. It is intended to extend its application to the characterization of the fascicles delimited by the IMCT. As a generic method, it will be adapted to magnetic resonance images at a higher scale on the same samples.

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## Figures and tables

Figure 1. Transversal sections of 4 bovine muscles (Biceps femoris (BF), Infraspinatus (IS), Longissimus dorsi (LD), Pectoralis profundus (PP)) stained with red Sirius





Figure 2. Reference maps of a transversal muscular section. a) original image, b) drawing of one judge, c) reference card which gives the probability of belonging to the class « perimysial network » d) segmentation of a fuzzy map for confidence level  $\varepsilon = 80\%$  and e)  $\varepsilon = 30\%$ 



Table 2. Correlation coefficients between the segmented area obtained by FCT and the collagen amount determined on the muscular sections for different confidence levels ( $\varepsilon$ )

E (%)	55	60	65	70	75	80
(R <sup>2</sup> )	0,10	0,44	0,60	0,71	0,76	0,80

Table 3. Connective network measures calculated from the		Collagen amount	Mean th (µ	nickness m)	Are (% total i	a image)	Len (mi	gth n)
bovine muscles. Different	Confidence level (ε)	(µg collagen /µg protein)	55%	80%	55%	80%	55%	80%
letters mean significant	BF	0.022 a	82 <sup>a</sup>	82 <sup>a</sup>	13,8 <sup>b</sup>	4,9 <sup>a</sup>	330 <sup>b</sup>	118 °
differences at $p=0.05$ per	IS	0.019 b	69 <sup>c</sup>	58 °	16,6 <sup>a</sup>	4,0 <sup>b</sup>	469 <sup>a</sup>	135 <sup>b</sup>
column.	LT	0.012 c	74 <sup>b</sup>	58 °	14,0 <sup>b</sup>	3,1 °	374 <sup>b</sup>	104 <sup>d</sup>
Mean thickness =area/length.	РР	0.022 a	80 <sup>a</sup>	67 <sup>b</sup>	14,8 <sup>ab</sup>	4,9 <sup>a</sup>	363 <sup>b</sup>	144 <sup>a</sup>



## EFFECTS OF PELVIC SUSPENSION ON THE TENDERNESS OF MEAT FROM FALLOW DEER (DAMA DAMA)

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#### Background

Texture, flavour and tenderness are attributes valued by consumers as very important in relation to the eating quality of meat. Different populations of consumers have different preferences for these quality attributes, something that affects the market for all types of meat. However, regardless of the consumer group, the consistency of meat quality is very important, and the product should be of the same quality every time it is purchased. In the Australian beef grading system Meat Standards Australia (MSA) these consumer important sensory quality attributes have been weighted in an overall score where tenderness represents 40%, flavour 20%, juiciness 10% and overall liking 30% (MSA, 2001).

Variation in meat tenderness and techniques developed to minimise this variation, have been investigated in several animal species over a long time period. However, there are very few studies performed on sensory quality attributes of deer meat (venison). Stevenson-Barry *et al* (1999) found a similar relationship between meat ultimate pH and tenderness of red deer (*Cervus elaphus*) venison (*M. longissimus*) to that reported for beef (Smulders *et al.*, 1990; Barnier, *et al.*, 1992). In contrast, reindeer (*Rangifer tarandus tarandus*) *M. longissimus* has been found to be extremely tender regardless of ultimate pH (Wiklund *et al.*, 1997). This phenomenon has been related to the speed of post mortem proteolysis (Wiklund *et al.*, 1997) and the small muscle fibre size (Taylor *et al.*, 2002) in reindeer.

It is well known that the conditions during rigor development (*e.g.* muscle pH decline, temperature/pH relationship and carcass treatment) are very important in controlling meat tenderisation (Dransfield, 1994). Therefore, carcass suspension techniques have been studied for beef (Hostetler *et al.*, 1970; Lundesjö Ahnström *et al.*, 2003) where the variation in tenderness is considered to be the main reason for consumer dissatisfaction (Koohmaraie, 1996). To our knowledge, the effect of pelvic suspension on tenderness in deer meat has not been previously studied.

## Objectives

The objective of this project was to study the effect of pelvic suspension on tenderness in fallow deer meat.

#### Materials and methods

Eight fallow deer bucks (18 months old, average live weight 42 kg, body condition score (BCS) 2-3 (Flesch, 2000), 7 fallow deer bucks ( $\geq$  36 months old, average live weight 57 kg, BCS 2-3) and 10 fallow deer does (24 months old, average live weight 38 kg, BCS 2-4) raised at the University of Western Sydney, were included in the study. The animals were fasted for 16 h prior to slaughter, stunned with a captive bolt and bled using thoracic stick exsanguination within 10 s of the stun (ethics approval UWS 00.09). The dressed carcasses were split along the mid ventral axis approx. 45 - 75 min post slaughter. The left side of each carcass was assigned to Achilles tendon suspension (control treatment) and the right side of each carcass was assigned to pelvic suspension. At 2 days post slaughter, 9 selected muscles were collected from each carcass-half (*Mm. semimembranosus, adductor femoris, biceps femoris, semitendinosus, vastus lateralis, rectus femoris, psoas major, longissimus* and *supra spinatus*). The meat samples were vacuum packaged, frozen and stored at -20° C until analysis. Meat samples were cooked on a Silex grill for 4-7 min at 240° C and then wrapped in aluminium foil and allowed to rest for 5 min. Internal temperature was measured during and after cooking to 60-65° C, which is equivalent to medium doneness according to the method described by Shaw (2000). Meat samples were cut to a 1 cm<sup>2</sup> core, with a minimum of 5 replicate sub-samples taken from each muscle for analysis. Meat tenderness was measured using a Warner Bratzler Shear force attachment on a



Stable Micro System TAXT2. Texture analysis was measured by means of force versus time in compression with a crosshead speed of 0.8 mm/s and a trigger force of 10 g with a contact area of 1 mm and contact force of 5 g to determine peak force. The data for each experiment was analysed statistically by residual maximum likelihood (Patterson & Thompson, 1971), with the random effects given by reading within muscle within animal, and the fixed effects by hanging treatment, muscle and their interaction, using the statistical package GenStat (2002).

## **Results and discussion**

The present results suggest that pelvic suspension of the carcasses had the greatest impact on meat tenderness in venison from the young male fallow deer (18 months old, Fig. 1), some impact on tenderness in venison from the older male deer ( $\geq$  36 months old, Fig. 2) but no significant impact at all on tenderness in venison from the female deer (24 months old, Fig 3). In studies of beef, similar differences in effect of pelvic suspension on meat tenderness for bulls and heifers have been reported. The tenderness in meat from bulls was more improved as an effect of pelvic suspension compared with meat from the heifers (Fisher, 1994; Lundesjö Ahnström *et al.*, 2003).

In the carcasses from the young fallow deer bucks, the tenderness of the following muscles was significantly improved ( $p \le 0.05$ ) as a result of pelvic suspension; *Mm. longissimus, biceps femoris, semimebranosus, adductor femoris* and *vastus lateralis*. These results are in good agreement with earlier studies on beef, where the tenderness of *Mm. longissimus, semimembranosus* and *adductor femoris* was positively affected by pelvic suspension (Hostetler *et al.*, 1970; Bouton *et al.*, 1973). For the older fallow deer bucks, significant effects of pelvic suspension on meat tenderness were found in *Mm. biceps femoris* and *semimembranosus*. The muscles that improved in tenderness as a result of pelvic suspension in the present study are all part of the most valuable cuts in a deer carcass; *M. longissimus* (striploin), *Mm. semimembranosus* and *adductor femoris* (topside), *M. biceps femoris* (silverside) and *M. vastus lateralis* (knuckle).

## Conclusions

The positive effect of pelvic suspension on tenderness in venison from the young male fallow deer is important information to consider for the Australian deer industry. This type of animal represents the deer most likely to be supplied for commercial slaughter in Australia. In addition, the important commercial cuts from female deer were generally more tender than the same cuts from males. The slaughter of female deer therefore provides a good option for farmers wishing to supply chilled venison year-round, especially at times of the year when the quality of venison from male deer is negatively affected by the breeding season.

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#### Fallow deer bucks, 18 months old

Figure 1. Shear force mean values in 7 muscles (LD = M. longissimus, BF = M. biceps femoris, ST = M. semitendinosus, SM = M. semimembranosus, AF = M. adductor femoris, VL = M. vastus lateralis and RF = M. rectus femoris) from fallow deer bucks (18 months old, n=8).





Figure 2. Shear force mean values in 9 muscles (SS = M. supraspinatus, PS = M. psoas major, LD = M. longissimus, BF = M. biceps femoris, ST = M. semitendinosus, SM = M. semimembranosus, AF = M. adductor femoris, VL = M. vastus lateralis and RF = M. rectus femoris) from fallow deer bucks ( $\geq$ 36 months old, n=7).



Figure 3. Shear force mean values in 9 muscles (SS = M. supraspinatus, PM = M. psoas major, LD = M. longissimus, BF = M. biceps femoris, ST = M. semitendinosus, SM = M. semimembranosus, AF = M. adductor femoris, VL = M. vastus lateralis and RF = M. rectus femoris) from fallow deer does (24 months old, n=10).



## EFFECTS OF HIGH OXYGEN PACKAGING ON TENDERNESS AND QUALITY CHARACTERISTICS OF BEEF *LONGISSIMUS* MUSCLES

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#### Background

High oxygen atmospheres, usually consisting of 70 - 80 % oxygen ( $O_2$ ) and 20 - 30 % carbon dioxide ( $CO_2$ ) are commonly used for retail meat packaging. The formation of bright red oxymyoglobin is beneficial, but pigment and lipid oxidation, premature browning of cooked meat, as well as microbiological spoilage, are more prone in high  $O_2$  than anaerobic environments (Hunt *et al.*, 1999; Sørheim *et al.*, 1999; Jakobsen and Bertelsen, 2000; Tørngren 2003). Recent findings indicate that display of beef in high  $O_2$  atmospheres negatively affects the development of tenderness of the meat (Tørngren, 2003). Little information exists on the extent and causes for the detrimental effect of high  $O_2$  packaging on tenderness. Aitch bone suspension from the *obturator foramen* is a well-known method for stretching pre-rigor muscles in beef carcass sides, thereby reducing muscle contraction and toughening (Hostetler *et al.*, 1972). If a detrimental effect of high  $O_2$  display on tenderness development is confirmed, it may be desirable to combine this packaging technology with a method like aitch bone suspension to ensure a satisfactory tenderness and quality of the meat.

## Objectives

To study the effects of aitch bone suspension and high  $O_2$  packaging on tenderness and other quality characteristics of beef *longissimus dorsi* (LD) muscles.

## Materials and methods

The experiment consisted of LD's from 4 treatments.

Nine bulls were assigned to treatments A and B:

A – aitch bone suspension + 14 days of high  $O_2$  display of steaks

B - aitch bone suspension + 14 days of vacuum display of steaks

Nine other bulls were assigned to treatment C and D:

C – aitch bone suspension + 14 days of high  $O_2$  display of steaks (equal to A)

D – traditional Achilles tendon suspension + 7 days of vacuum storage of whole muscles + 7 days of high  $O_2$  display of steaks.

The eighteen bulls were of different breeds and were sampled randomly at a commercial abattoir. The average carcass weight was 349 kg within a range of 264-426 kg. The carcasses were electrically stimulated with low voltage. The sides of the carcasses were assigned to two treatments with alternate left and right sides for each treatment. Aitch bone suspension was performed within 60 minutes p.m. The sides were chilled at an air temperature of appr. 4 °C to an average LD core temperature of 11 °C at 10 hours p.m. Two days after slaughter the sides were deboned, and the LD's were cut in steaks 15 mm thick, except 35 mm for Warner – Bratzler (WB) analysis. Steaks were packaged either in 70 %  $O_2/$  30 %  $CO_2$  in trays on a Ross Cryovac tray top-seal machine (Ross Cryovac) with film and tray oxygen transmission rates of appr. 15 cm<sup>3</sup>/m<sup>2</sup>/24 h. at 23 °C and 0 % RH, or in vacuum on an Intevac IN30 chamber machine (Intevac Verpackungsmaschinen, Wallenhorst, Germany) with polyamide bags with oxygen transmission rate of 40 cm<sup>3</sup>/m<sup>2</sup>/24 h. at 23 °C and 75 % RH. Meat for all analyses was packaged, except unpackaged meat for the first WB and sensory analyses at 3 days p.m. The meat was displayed in darkness at 4 °C for 15 days.

The following analyses were performed at 3, 10 and 17 days p.m.:



- Warner-Bratzler peak shear force was measured on samples heated in a circulating water bath at 70 °C for 50 min. to a core temperature of 69.5 °C and cut in strips of 10 x 10 x 35 mm with 10 replicates per sample (not at 10 days p.m.)
- Sensory analysis was performed with 11 trained assessors with a quality descriptive test (ISO 6564) for tenderness, hardness, juiciness, rancid taste and rancid odour on samples heated at 70 °C for 40 min. in a water bath to 70 °C core temperature with evaluation in red light for masking possible differences in premature browning (Hunt *et al.*, 1999)
- L\*a\*b\* (lightness, redness, yellowness) was analysed with a Minolta Chroma Meter CR-300 (Minolta Camera Co., Osaka, Japan) with 8 mm viewing port and illuminant D<sub>65</sub> with 3 replicate measurements per sample (treatment D was not analysed at 3 days p.m.)

In the statistical methods, the emphasis was on comparing the effect of display time separately for each treatment, as opposed to comparing the treatments. Analysis of variance (ANOVA) was used for the analyses, with Tukey's Multiple Comparison Test to determine in detail which groups or display times that were different (SAS System Release 8.2, SAS Institute Inc., Cary, NC, USA).

## **Results and discussion**

Table 1.Warner-Bratzler peak shear force (N/cm<sup>2</sup>) and standard deviation (+/-) of LD steaks during display at  $4 \degree C$ .

Time,		Treatment								
days p.m.	Α	В	С	D						
3	59.9 a +/- 11.7	59.9 a +/- 11.7	63.0 a +/- 17.9	72.7 a +/- 25.6						
17	54.7 a +/- 9.6	47.7 b +/- 7.1	57.5 a +/- 7.1	65.2 a +/- 15.8						
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For description of treatments, see materials and methods

a,b. Means in the same column with different letters are significantly different (p < 0.05).

Table 2. Sensory analysis of tenderness, hardness and juiciness of LD steaks during display at 4 °C. Scale: 1 = none, 9 = very much.

Time,	Tenderness					Hardn	ess		
days p.m.		Treatme	ent	Treatment					
	А	В	С	D	Α	В	С	D	
3	4.2 a	4.3 b	4.1 b	3.5 b	5.5 a	5.4 a	5.5 a	6.1 a	
10	4.7 a	5.8 a	4.7 ab	5.2 a	5.1 a	4.2 b	5.0 a	4.8 b	
17	4.9 a	5.8 a	5.0 a	4.8 a	4.9 a	4.1 b	4.8 a	4.9 b	
		Juiciness							
		Treatmen	nt						
	А	В	С	D					
3	4.8 a	4.8 a	5.2 a	5.3 a					
10	3.9 b	4.8 a	4.1 b	4.9 a					
17	3.9 b	4.2 a	4.2 b	4.2 b					

For description of treatments, see materials and methods

a,b. Means in the same column with different letters are significantly different (p < 0.05).

Warner-Bratzler shear force (Table 1) and sensory tenderness and hardness values (Table 2) showed that treatment B with 14 days of vacuum display improved the tenderness of the meat (p < 0.05), but particularly during the first 7 days, as demonstrated by the sensory analysis. Over all, treatment B with a combination of aitch bone suspension and vacuum display gave the most tender meat. Meat of treatment A, with 14 days of high O<sub>2</sub> display, did not significantly change in shear force, tenderness and hardness during the display period (p > 0.05); neither did meat of treatment C change in shear force and hardness (p > 0.05). Meat of treatment D with 7 days of vacuum, followed by 7 days of high O<sub>2</sub> display, significantly increased in tenderness and decreased in hardness during the first 7 days of vacuum (p < 0.05), but did not change in tenderness or hardness during the subsequent high O<sub>2</sub> display (p > 0.05). Steaks of treatments A and C in high O<sub>2</sub> lost juiciness after 7 days display (p < 0.05), as well as steaks from treatment D between days 10 and 17 p.m. Vacuum packaging did not alter juiciness during display, either for treatment B or D (p > 0.05). The lack of tenderisation by high O<sub>2</sub> treatments is in support to a previous study by Tørngren (2003), who found that beef LD steaks displayed for 16 days in 80 % O<sub>2</sub> / 20 % CO<sub>2</sub> were less tender than steaks displayed in



vacuum or 50 %  $CO_2/$  50 %  $N_2$ . In a comparison of 100 %  $CO_2$  and vacuum storage of beef, no differences in tenderness were found during storage (Bell *et al.*, 1996), making it likely that  $O_2$ , and not  $CO_2$ , is the gas responsible for inadequate tenderisation.

Before display, meat assigned to all treatments was relatively tender, due to relatively high carcass weights, use of electrical stimulation and relatively slow chilling. Although meat treated with aitch bone suspension tended to be slightly more tender than meat from traditional Achilles tendon suspension at start of packaging, no significant differences were found in average peak force between treatments C and D (p > 0.05). However, aitch bone suspension reduced the standard deviation of the WB shear force within the group by both day 3 and 17 p.m. compared to Achilles tendon suspension, particularly by lower force of LD's from the toughest carcasses. Previous studies showed that aitch bone suspension of beef and veal carcasses was significantly beneficial to tenderness at conditions inducing cold shortening, but to various degrees at slow chilling rates (Sørheim *et al.*, 2001; Wahlgren and Kalbakk, 2002; Wahlgren *et al.*, 2002). In these studies, aitch bone suspension also reduced the variation in WB shear force by improving tenderness of the toughest meat.

Table 3. Sensory analysis of rancid taste and odour of LD steaks during display at 4 °C. Scale: 1 = none, 9 = very much.

Time,		Rano	cid		Rancid				
days p.m.		taste odour							
		Treatme	ent	Treatment					
	А	В	С	D	А	В	С	D	
3	1.1 b	1.1 b	1.1 b	1.0 b	1.1 b	1.1 b	1.1 b	1.1 b	
10	2.9 a	1.1 b	2.3 a	1.1 b	1.8 ab	1.3 b	1.9 a	1.3 ab	
17	3.5 a	2.5 a	2.4 a	2.4 a	2.4 a	2.5 a	2.0 a	2.1 a	

For description of treatments, see materials and methods.

a,b. Means in the same column with different letters are significantly different (p < 0.05)

High  $O_2$  display increased rate of development of rancid taste in the steaks (Table 3), as shown for treatment A and C as early as 7 days display, and for treatment D at 14 days of display (p < 0.05). An increased rancid odour developed in meat of treatment C after 7 days display (p < 0.05). The findings are in agreement with Jakobsen and Bertelsen (2000), where 55 – 80 %  $O_2$  storage of beef LD steaks at 4 - 8 °C caused lipid oxidation.

Time,	a*				b*			
days p.m.	. Treatment Treatme						ent	
	А	В	С	D	А	В	С	D
3	25.9 a	19.4 a	26.2 a		14.3 a	5.6 a	14.0 a	
10	22.4 b	19.0 a	21.9 b	26.3 a	12.9 b	5.4 a	12.0 b	13.6 a
17	19.7 c	18.5 a	20.0 c	23.1 b	12.3 b	5.4 a	11.8 b	12.4 a

Table 4. a\* (redness) and b\* (yellowness) values of LD steaks during display at 4 °C.

For description of treatments, see materials and methods.

a,b,c. Means in the same coloum with different letters are significantly different (p < 0.05)

Packaging conditions affected meat colour (Table 4). a\* redness values of the high  $O_2$  treatments A, C and D decreased during display, but not below the a\* value of the vacuum treatment B at 14 days. b\* yellowness values were considerably higher for all  $O_2$  treatments than the vacuum treatment at all sampling times (p < 0.05). L\* lightness values were higher for treatment A than B at all sampling times (p < 0.05), but treatment C and D did not differ in L\* values (p > 0.05) (results not shown). Although some redness was lost from high  $O_2$  samples during the 14 days display, the meat was still fairly red and not discoloured at end of display. Vacuum samples had stable L\*, a\* and b\* values at 3, 10 and 17 days p.m.

## Conclusions

The study confirmed previous indications of reduced tenderness development in beef LD steaks displayed in high  $O_2$  atmospheres. In addition, steaks displayed in high  $O_2$  were less juicy. Steaks in vacuum improved in tenderness during the display period, and these steaks were ranked highest in final tenderness. The extent and



causes of the negative effect of high  $O_2$  packaging on tenderness, as well as precautionary measures to reduce or avoid this effect, need to be addressed further. Aitch bone suspension did not significantly improve tenderness, probably because the meat was relatively tender by other tenderizing measures in the experiment, but the variation in tenderness was lower with aitch bone than Achilles tendon suspension. High  $O_2$  display increased rancidity of the meat. The initial bright red colour decreased during high  $O_2$  display, but not to the extent of discolouration.

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