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Extending the shelf life of lamb meat

The effects of medium voltage electrical stimulation, CO₂ ageing and vitamin E supplementation on the colour stability of lamb meat

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1. Executive summary

This project consisted of four experiments to investigate the effects of recent processing improvements on lamb meat colour stability.

- Experiment 1 The effect of vitamin E, supplied through supplements or green pasture, on colour stability of lamb meat that has not been electrically stimulated
- Experiment 2 The effects of medium voltage electrical stimulation, ageing and vitamin E supplementation on the colour stability of lamb meat
- Experiment 3 Consumer benchmarks for colour stability
- Experiment 4 Comparison of different Hunterlab spectrophotometers for the purpose of measuring meat colour stability

At the completion of the experimental work, recommendations were synthesised from data obtained from the different experiments into a summary document for the purpose of consultation with industry members ("Commercialising Lamb Meat Colour" Appendix 2).

- Packaging meat as primal cuts in carbon dioxide for 10 days produced a highly acceptable colour and colour stability for lamb meat.
- Medium voltage electrical stimulation caused no detrimental effects on retail display time under the current 48h shelf life scenario.
- Results indicated that lamb meat should not be aged for longer than 10 days unless the vitamin E status of the lambs prior to slaughter can be assured. Extended ageing to 30 days is likely to reduce colour stability to an unacceptable level when the vitamin E concentration in meat is low.
- Supplementation of the lamb finishing diet with vitamin E can improve the colour stability of lamb meat. For lambs backgrounded on dry pasture, the recommended rate of inclusion of vitamin E in a feedlot diet is 250ppm. This should be fed for a minimum period of 2 weeks prior to slaughter.
- To ensure that meat is attractive to consumers, the oxy/met ratio of lamb meat should not fall below 3.5 during the retail display period.
- Display period could be extended from 48h to 60h but would involve consideration of cut, lamb history, and ageing time.

2. Background

2.1. Introduction

In recent years Woolworth's lamb business has undergone significant change with carcases receiving medium voltage electrical stimulation (MVS) and primal cuts are subsequently 'aged' under a CO₂ gas flushed system. The lamb meat is then dispatched to stores where it is sliced and displayed in the usual over wrap manner. This major change in the lamb meat business created a need to clarify the effects of electrical stimulation and ageing on retail colour display determined by metmyoglobin formation. Meat colour stability is a major issue for meat retailers. Currently the value of the discount applied to meat that is on shelf for 48 hours is estimated to be \$4million to Woolworths per week nationwide. Not all of this can be attributed to colour stability but the 48 hour benchmark for discounting meat is based on colour stability.

2.1.1. Effects of cut and aging

Previous research by the sheep meat eating quality program has highlighted the effects of cut (Table 1) and days aging under vacuum (Table 2). Clearly leg cuts are more unstable than the loin.

Cut	% packs sc	ored ≥3 for retail a	acceptability
- Cui	3d	5d	9d
Loin	88	27	0
Knuckle	61	0	0
P value	<0.001	<0.001	<0.001

Table 1. Retail colour acceptability of over wrapped packaged lamb after 3, 5 & 9 days
retail display (adapted from Channon et al (2004))

Aging past 21 days in a vacuum bag was associated with a marked decline in colour stability (Table 2).

Primal	Days in vacuum	% packs scored ≥3 fo	or retail acceptability
	5	3d	5d
	7	100	80
Loin	21	100	0
	35	63	0
	P value	P<0.001	P<0.001
	7	83	0
Knuckle	21	78	0
	35	22	0
	P value	P<0.001	P<0.001

Table 2 Effect of ageing time in vacuum (5, 21 & 35 days) on the retail acceptability of MAP and over wrapped packaged lamb (adapted from Channon et al (2004))

2.1.2. Electrical stimulation

Electrical stimulation is rapidly being adopted by lamb processors in Australia (2005) to reduce variability in eating quality. Jose (2004) confirmed the findings of other studies that electrical stimulation can improve the initial colour in chilled lamb meat without any negative effect on colour stability. However, Moore and Young (1991) found that the effect of electrical stimulation on meat colour and colour stability depended on factors such as ageing and freezing.

Electrical stimulation systems vary from low to medium and high voltage. The new systems being adopted in Australia are medium voltage and the effect of these systems on colour stability is unclear. Jacob *et al* (2007) found a small and inconsistent effect of electrical stimulation with a high voltage system that depended on lamb age and cut.

2.1.3. Effects of Vitamin E

Another area affecting retail colour stability is the feeding history of the lamb before slaughter. Vitamin E is a powerful antioxidant that is thought to prevent oxidation of myoglobin to metmyoglobin both directly and indirectly by preventing the oxidation of phospholipids in cell membranes (Faustman *et al.*, 1998). In addition to

improvements in colour stability, reductions in drip loss and improvements in flavour of aged product have been reported in response to vitamin E supplementation (Mitsumoto *et al.*, 1995). These latter effects have been attributed to improvements in cell membrane integrity and reductions in rancidity flavours due to lipid oxidation post mortem. However the potential value of vitamin E supplementation to improve the colour stability of lamb meat in Australia has to date largely been unaccounted for.

Reports in the literature on the beneficial effects of vitamin E supplementation for extending the shelf-life of meat mostly relate to cattle although there have been some studies done with sheep (Wulf *et al.*, 1995; Turner *et al.*, 2002). In Western Australia, Pearce and Jacob (2005) demonstrated that meat from Merino lambs grazing saltbush had improved colour stability compared to meat from Merino lambs grazing dry stubble and attributed this effect to a difference in vitamin E concentration between the diets

Nutritional myopathy, a debilitating Vitamin E deficiency syndrome is endemic in young sheep in the south west of WA and occurs when the Vitamin E concentration in muscle falls below 1.5 mg/kg (GM Smith pers comm.). Reserves in muscle become depleted when the diet is low in vitamin E for about 4 weeks (Fry *et al.*, 1993) hence the prevalence of nutritional myopathy in the summer autumn period since Vitamin E is high in 'green' feed. The effect of low vitamin E concentration on meat colour stability are likely to be widespread because the concentration required to optimise meat colour stability is considerably higher (about 3mg/kg) than that required to prevent nutritional myopathy. Preliminary studies suggest that vitamin E concentrations in muscle of less than 2mg/kg are common in prime lambs during the summer and autumn period. This background data fits with observations that shelf life is low for lambs sourced during the autumn and early winter months. Other nutritional factors may interact with vitamin E concentration particularly the concentration of pro-oxidants such as polyunsaturated fatty acids that vary according to feed type.

2.1.4. Consumer perception of meat colour stability

The perception of freshness and wholesomeness by consumers is governed by meat colour (Young *et al.*, 1999; Mancini & Hunt, 2005) with consumers preferring meat to be red rather than brown. Metmyoglobin formation, the cause of meat browning, can be assessed by measuring light reflectance at two wavelengths (580nm and 630nm) and comparing the ratio of these values (Hunt, 1991). This ratio is known as the

oxy/met ratio. However, relatively little is known about the relationship between this objective measure and the perception of meat colour by consumers. Evaluating the success of interventions to prevent browning depends on the consumer perception of colour change during shelf display, and being able to relate objective measurements to consumer perception.

3. Project objectives

This project aimed to investigate the impact of some new meat processing technologies, namely medium voltage electrical stimulation and CO_2 gas flushing of primal cuts, on the colour stability of meat obtained from lambs finished with different feed types and vitamin E status. The purpose of the project was to understand the effects of these recent processing changes to the quality of meat produced by the Woolworth's supply chain. In addition to this, a further aim was to understand how to increase shelf time from 48h to 60h, on a commercial cuts basis, within the context of the entire supply chain from farm to retail shelf.

4. Methodology

4.1. Experiment 1 The effect of vitamin E, supplied through supplements or green pasture, on colour stability of lamb meat that has not been electrically stimulated

4.1.1. Lambs and dietary treatments

Weaned first cross Poll Dorset Merino lambs of mixed gender were used for the experiment. The lambs were purchased from a commercial farming property at Toodyay W.A. and transported to the Murdoch University farm for the experiment which began in March 2005.

A preliminary group of 10 lambs (dry feed group) were slaughtered immediately. These lambs came directly from the farm of origin at Toodyay and had been supplemented with 600grams of lupins per head per day on a senesced annual ryegrass pasture.

The remaining lambs were drenched with Cydectin© anthelmintic that contained selenium and then allocated to one of the following finishing treatments:

- 1. Pellet group 1- with 30 IU vitamin E /kg DM for 56 days (control),
- 2. Pellet group 2- with 150 IU vitamin E /kg DM for 56 days,
- 3. Pellet group 3- 275 IU vitamin E /kg DM for 56 days,
- 4. Pellet group 4- 400 IU vitamin E /kg DM for 56 days,
- Green pasture- a mixed sward irrigated pasture (kikuyu/clover/ryegrass) for 56 days. The clover contained 97mg/kg of vitamin E, the kikuyu grass pasture 127 mg/kg on a dry matter basis and no estimate was made for the ryegrass.

For each of these treatments there were 2 replicates of 6 lambs giving a total of 60 lambs. After slaughter each carcass was cut in 2 halves. One half was cut into primal cuts, the bones removed from front and hind legs and packed loosely in plastic until cut for shelf display 5 days after slaughter (fresh pack). The other half was prepared similarly but packed in CO_2 and cut for display 21 days after slaughter (gas pack). All cuts were stored at $2^{\circ}C$. The bones were left in for the loin primal cut. Lambs were slaughtered at the V&V Walsh abattoir, boned and packed at the

Woolworths meat centre Bunbury. Then primal cuts were cut for display at Murdoch University Meat Science Laboratory.

4.1.2. Meat preparation and display

Individual muscles were dissected and cut transversely into slices 3 cm in thickness for display. These muscles included *m. longissimus thoracis et lumborum* (short loin), *m. semimembranosus* (topside), *m. semitendinosus* (eye round), *m. rectus femoris* (round), *m. gluteus medias* (rump), and *m. triceps brachii* (shoulder).

For shelf display individual muscles were over wrapped with PVC cling wrap on black polystyrene foam trays and kept in a continuously lit drinks display refrigerator, at 5^oC for 4 days, at Murdoch university meat science laboratory.

4.1.3. Measurements

During the feeding period blood samples, muscle biopsies and live weights were collected fortnightly. The muscle biopsies were taken from the *m. semimembranosus* and *m. semitendinosus* using the method described by Gardner *et al* (2001). After the lambs were slaughtered samples were taken from liver and muscles for vitamin E analyses.

During shelf display colour measurements including L, a, b and reflectance at light wavelengths 580nm and 630 nm were recorded with a Hunterlab 45O/L reflectometer 30 minutes after the meat had been cut. Hue, chroma, oxy/met and met formation were subsequently calculated from these measurements. Measurements were done at 0, 6, 18, 30, 42, 54, 66, 78, 90 and 96 hours after cutting and over wrapping. Samples were prepared for drip loss measurement at the time of cutting for shelf display and pH was also measured at this time to estimate ultimate pH (pHu).

Vitamin E (α -tocopherol) concentrations were measured by the Animal Health Laboratory, Department of Agriculture and Food WA, South Perth, WA. Alpha-tocopherol in plasma, muscle, liver and feed was measured using high performance liquid chromatography (HPLC) with fluorescence detection (McMurray & Blanchflower, 1979). Tissues and feed were saponified using the method of Bieri *et al* (1961) before extraction with hexane.

4.1.4. Statistical analyses

Colour data was analysed using a linear mixed effects model with display time as a covariate, diet and packaging type as fixed effects, and animal as a random term (SAS[®]). The oxy/met ratio at 48 and 60 hours for each individual was predicted by fitting an exponential equation. Linear mixed effects models (SAS[®]) were used to test muscle vitamin E concentration as a covariate on the predicted oxy/met ratios, with animal as a random effect.

4.2. Experiment 2 The effects of medium voltage electrical stimulation, ageing and vitamin E supplementation on the colour stability of lamb meat

4.2.1. Lambs

Weaned, first cross Poll Dorset Merino ewe lambs, born in July 2005 and shorn in October 2005, were used for this experiment. These lambs were purchased from the same commercial farming property at Toodyay that supplied lambs for experiment 1, and relocated to the Murdoch University veterinary farm on December 19 2005. Prior to purchase the lambs had been grazed on dry senesced annual ryegrass pasture. The lambs were drenched with Cydectin© anthelmintic that contained selenium and introduced to a pelleted finishing diet on arrival at Murdoch University. The experimental diets were fed for a period of 5 weeks commencing on January 2 2006. The lambs were then slaughtered as one consignment at the V&V Walsh abattoir Bunbury on February 7 2006.

4.2.2. Experimental design

The experiment design was a 2X2 factorial design for vitamin E (n=40) and MVES (n=40) with 4 ageing treatments nested within vitamin E and MVES treatments. The vitamin E treatments were; no added vitamin E (control) and vitamin E (α -tocopherol acetate) at the rate of 250 ppm (VitE). The MVES treatments were no stimulation (NES) and stimulation (ES), and the ageing treatments were ; 5 d (D5), 10 d (D10), 20 d (D20) and 30 d (D30). The vitamin E addition rate of 250ppm was derived from blood, muscle and meat colour results from experiment 1.

Electrical stimulation was applied with a commercially installed post dressing MVES unit that applied electrical stimulation for 30 seconds via rubbing bars at the level of the shoulder and hocks. A setting of 1.2mS pulse duration, 0.8A current and 14hz frequency was used.

Carcasses were cut into primal cuts 24 h approximately after slaughter. Sides were allocated to ageing treatments such that equal numbers of left and right sides were allocated to each treatment group. The primal cuts were then stored at a temperature of 2°C in dark conditions until cutting for display at a time appropriate for each ageing period. Primal cuts aged for 5 days were packed loosely in plastic bags that allowed exposure to air, to represent fresh carcass trade meat. Primal cuts aged for 10, 20 and 30 days were packed in sealed plastic bags that had been flushed with a gas containing 100% carbon dioxide, to represent the current system used by the Woolworths supply chain.

4.2.3. Meat preparation and display

Individual muscles were dissected and cut transversely into slices 3 cm in thickness for display. The muscles prepared in this way were *m. longissimus thoracis et lumborum* (short loin), *m. semimembranosus* (topside), and *m. semitendinosus* (silverside).

The sliced meat was placed on black polystyrene foam trays, over wrapped with PVC cling wrap, and kept in a continuously lit drinks display refrigerator, at 3^oC for 4 days, at Murdoch university meat science laboratory.



Figure 1 Allocation of treatments

4.2.4. Measurements

Blood samples and live weights were collected from live lambs every 14 days of the feeding period for vitamin E analyses. After the lambs were slaughtered samples were taken from liver and muscles for vitamin E analyses.

During shelf display, colour measurements including L, a, b and reflectance at light wavelengths 580nm and 630 nm were recorded with a Hunterlab reflectometer. Hue, chroma, oxy/met and met formation were subsequently calculated from these measurements. Measurements were done at 0, 6, 18, 30, 42, 54, 66, 78, 90 and 96 h after cutting and over wrapping. The measurement at 0h was done at least 1h after slicing to allow the meat to "bloom". Samples were prepared for drip loss measurement at the time of cutting for shelf display and pH was also measured at this time to estimate ultimate pH (pHu).

4.2.5. Statistical analyses

Colour data was analysed using a linear mixed effects model with display time as a covariate, diet and packaging type as fixed effects, and animal as a random term (SAS[®]). In a separate analysis,

4.3. Experiment 3 Consumer benchmarks for colour stability

4.3.1. Consumers

A face to face survey was conducted in August 2006 with people shopping at the "The Captain Stirling" IGA store, Nedlands, Perth Western Australia. Ninety-one percent of consumers interviewed ate beef at least once a week while 82% of consumers ate lamb at least once a fortnight. Seventy-six percent of participants were from the Curtin federal electorate. Seventy-five percent of participants were female and 70% of participants had a university education (Table 2).

Factor	Category	Percentage
	20 – 29	34
Age (vears)	30 – 49	20
	50 – 69	31
	70+	15
Gender	Male	25
	Female	75
	High school / TAFE	27
Education	University	70
	Other	3

Table 3 Demographic description of the survey participants

4.3.2. Questionnaire

The questionnaire (Appendix 1) was composed of three parts. The first was background on how frequently respondents ate beef and lamb and demographics including: age, gender, postcode and education. The second section asked participants to view a meat tray containing eight beef samples varying in colour from red to brown. Participants were asked to view each sample of beef provided and place a vertical line at the point on the colour scale (red to brown) that they thought corresponded to the colour of the sample. Participants then placed a tick in the adjacent box if they would be happy to eat the sample of meat or a cross if they would not prefer to eat it. Participants repeated this process for the second tray of meat containing eight samples of lamb varying in colour from red to brown. The final section consisted of questions to determine participants' attitudes towards red meat colour. The first question offered a 'yes' or 'no' answer while the following nine questions used a six point scale of *strongly agree, agree, indifferent, disagree, strongly disagree* and *don't know*. There was also allocated space for comments if people wished to comment.

The questionnaire was pre-tested before interviews were conducted. Pre-testing of the questionnaire was important to ensure that the survey could be completed within five minutes, that instructions were clear with minimal confusion and that participants interpreted the questions in the way they were intended so the desired outcomes could be gained.

Interviews were conducted on a Tuesday morning (9am – 12pm), Wednesday afternoon (12.30 – 3.30pm) and Monday afternoon (1 - 4pm). Bias was minimised as much as possible with the same three interviewers, who were final year students from the University of Western Australia, present at all sessions and the questionnaire stand and meat were all located in the same position for all sessions. The interviews were conducted in the meat aisle towards the end to minimise interference with shoppers' activities. People who were under the age of eighteen, who had children or were obviously in a rush, were not selected for interview.

4.3.3. Meat

Loin (*m. longissimus*; LD) and topside (*m. semimembranosus*; SM) cuts of beef and lamb were used in the survey. Meat samples were cut at various stages throughout the week before the questionnaire to ensure samples displayed a range of colours from red through to brown. It was anticipated that colour (oxy/met) would be consistent for each survey day. Samples were uniformly cut with no exterior fat. Eight approximately 3 x 2 cm cubes of beef, four loin and four topside, varying in colour from red to brown were randomly placed in 2 x 4 format on a black meat tray wrapped with cling wrap. The tray was labelled BEEF and labels from 1 to 8 were placed below each sample. The lamb samples were prepared in accordance with the beef samples. The tray was labelled LAMB and labels from 9 to16 were placed below each sample. The two meat trays were placed in the corner on the bottom shelf near the front of the meat cabinet (Tyler Model D2TM/12), under pink fluorescent lighting (NEC TRI – PHOSPHOR 37 watts, FL40SSBR –B/37/HG).

4.3.4. Colour measurement

The colour of the meat was measured, using a Hunter lab mini scan TM XE Plus 45/0 Large. Measurements were taken before and after each viewing session, and the mean of these two measurements was used to determine oxy/met ratio at the time of viewing. The oxy/met ratio was calculated by dividing the percentage reflectance of light at wavelength of 630nm by the percentage reflectance of light at a wavelength of 580nm.

4.3.5. Statistical analyses

The consumer colour score was calculated as a proportion of the distance the mark placed by respondents was from the red end of the colour scale. The scale was 5 cm in length so the distance was converted to a point on a continuous scale from 0 (brown) to a 100 (red) by taking the difference from 5 and multiplying by 20. A

logistic function was fitted to raw oxy/met ratio and colour perception data. The percentage of people who would eat the meat samples were calculated along with the percentages for all questions in the first and final sections. Four questionnaires were omitted from the data for question 7 and one questionnaire was omitted for question 9 as the questions were incorrectly completed. The statistical package Genstat was used for all statistical analyses (<u>http://www.vsn-intl.com</u>). Significance was determined by P-value <0.05. Over the three interview days sixty-seven completed questionnaires were collected with seventeen completed on day one, twenty-three on day two and twenty-seven on day three.

4.4. Experiment 4 Comparison of different Hunterlab spectrophotometers for the purpose of measuring meat colour stability

4.4.1. Spectrophotometers

This experiment was conducted at the Meat Science Laboratory DPI Victoria Werribee on November 21 2006. Three different Hunterlab miniscan TM XE Plus spectrophotometers identified as WA, VIC and NSW were evaluated. WA (model No. 45/0-L) had a large reading head (25mm) whilst both Vic and NSW had small (5mm) reading heads (model No. 45/0-S). The light source was set at "C" and the aperture set to 10. The instruments were calibrated on a white tile and black glass as directed by the manufacturer's specifications.

4.4.2. Experimental design

Sample of chicken breast (*m. pectoralis major*), pork loin (*m. longissimus thoracis et lumborum*), beef rump (*m. gluteus medias*) beef fillet (m. psoas major), mutton loin (*m. longissimus thoracis et lumborum*) and lamb topside (*m. semimembranosus*) were measured at the one measuring period. Meat samples were sliced to a thickness of 20 mm. The beef and lamb meat samples were cut at 0, 1, 4 and 7 days prior to measurement and displayed for the corresponding times in a meat display cabinet. The pork and chicken samples were cut and measured 30 minutes after cutting to allowing blooming on day 0 only. The tiles measured were high gloss ceramic domestic kitchen style tiles.

4.4.3. Meat display

An upright retail display cabinet with 4 adjustable shelves (Shelley Open Display, dimensions 1960 × 1890 × 823 mm, Quirk's Refrigeration Pty Ltd, Australia)

illuminated with white fluorescent lighting (36W, 3300 lm) along the front side walls was used to simulate retail display. Meat was packed on black polyurethane trays and over wrapped with oxygen permeable polyvinyl wrap.

4.4.4. Statistical analyses

A linear mixed model (REML) was used to test the effects of species, muscle, and spectrophotometer and display time on colour. The statistical package Genstat was used for all statistical analyses (<u>http://www.vsn-intl.com</u>). Significance was determined by P-value <0.05.

5. Results and Discussion

The results from the different experiments have been presented under headings that relate to the projects objectives rather than to be organised within experimental groupings. For details of the methods the reader needs to refer back to the methods section.

5.1. Cuts

Muscle and therefore commercial cut was seen to have a main effect on colour colour and colour stability. The different muscles varied in colour at the beginning of the display period and the rates at which they changed colour during display. The colour of meat when first cut (bloom colour) depended on the muscle both for lightness and hue (Figure 2 and 3). Topside had the lowest L value (the darkest muscle) followed by the loin, rump, round, shoulder and the silverside (eye of silverside) which was the lightest.

Figure 2 Lightness (L value) of different muscles at the time of cutting



L value at time of cutting

The loin had the lowest hue (was the most red in colour) and the silverside (the most brown in colour) of all the muscles at the time of cutting (Figure 3).







Muscles could be ranked according to the amount of colour change that occurred during display (Figure 4). From experiment 1 the ranking from most to least colour change was SM > GM > LL > TB > RF > ST. The more oxidative a muscle the unstable was the colour of the muscle. In relation to cuts this ranking for colour stability translates to topside > rump > loin > shoulder > knuckle > eye of silverside. This result was as expected except for knuckle that previously (Channon *et al.*, 2004) has been scored low for colour stability (Table 1).

Figure 4 The change in oxy/met ratio of different muscles with shelf display time (experiment 1, 5 day aged in air packaging)



5.2. Electrical stimulation

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Electrical stimulation had little effect on bloom colour. L and a values were not changed by electrical stimulation but b values were (Figure 5). Stimulated meat was therefore likely to appear redder at the beginning of the display period than non stimulated meat.

Figure 5 The effect of electrical stimulation on b value at the commencement of the display period (experiment 2)



There was no main effect of electrical on the rate of change in oxy/met ratio (Figure 6).



Figure 6 The effect of electrical stimulation on the change of oxy/met with time for different ageing periods (values are predicted means for all muscles experiment 2)

5.3. Ageing

Ageing reduced colour stability (Figure 7) although this effect depended on muscle and dietary treatment.

Figure 7 Predicted rate of change in oxy/met ratio during shelf display for 5, 10, 20 and 30 day aged meat (experiment 2, control diet and no electrical stimulation).



5.4. Vitamin E

Positive effects of vitamin E supplementation on colour stability were seen in experiments 1 and 2. Whilst this effect depended on the ageing period and the muscle, vitamin E supplementation generally delayed the onset of browning and increased potential shelf life.

5.4.1. The requirement for vitamin E

The different rates of supplementation used in Experiment 1 enabled a comparison between colour stability and vitamin E concentrations in muscle. There was a significant correlation (P<0.001) between vitamin E concentration in muscle and the rate of colour stability, expressed as the oxy/met ratio reached after 48h shelf life, when meat was aged for 30 days and no correlation when meat was aged for 5 days. Importantly the nature of this relationship suggested that once the minimum requirement for vitamin E has been reached there is no further improvement in colour stability if the muscle concentration is increased beyond this level. When meat was aged for 30 days the oxy/met at 48h shelf life increased until a threshold vitamin E concentration of about 3-4mg/kg was reached (Figure 8). This suggests that vitamin E concentration in lamb meat should exceed 3mg/kg to ensure good shelf life.



Figure 8 The effect of muscle vitamin E concentration on colour stability for 5 day air packed and 30 day CO_2 packed meat (experiment 1)

	Muscle					
Diet	GM	LD	RF	SM	ST	ТВ
Dry pasture	1.64±0.05 ^a	1.31±0.05 ^b	1.58±0.05 ^a	1.86±0.05 ^c	1.36±0.05 ^b	na
Pellet						
30 ppm vit E	2.02±0.26 ^a	2.12±0.18 ^ª	1.87±0.26 ^a	2.38±0.18 ^a	1.93±0.18 ^ª	2.14±0.26 ^a
Pellet						
150 ppm vit E	4.1±0.24 ^{ac}	4.04±0.18 ^{ac}	3.63±0.24 ^{ab}	4.35±0.18 ^c	3.51±0.18 ^b	3.98±0.24 ^a
Pellet						
275ppm vit E	4.81±0.24 ^a	4.79±0.18 ^a	4.5±0.26 ^{ab}	5.07±0.18 ^a	4.12±0.18 ^b	4.6±0.25 ^{ab}
Pellet						
400 ppm vit E	5.99±0.24 ^a	5.09±0.17 ^b	5.58±0.25 ^{ab}	5.48±0.17 ^{ab}	4.6±0.17 ^c	5.54±0.25 ^{ab}
Green pasture	4.24±0.26 ^{ab}	3.68±0.18 ^{ac}	4.35±0.26 ^b	3.98±0.18 ^{ab}	3.35±0.18 ^c	4.15±0.26 ^{ab}

Table 4 Vitamin	E concentration	in muscles	5 days post	t slaughter
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Values are means ±SEM, values with different superscripts within the same row are different

There was a significant (P<.001) effect of muscle, diet and packing system on vitamin E concentration post slaughter (Table 4). The concentration of vitamin E increased with increasing concentrations of vitamin E in the diet. The major effect of muscle was that the average concentrations of vitamin E across diets and packing systems were significantly lower for the ST (eye round) than for all other muscles. Otherwise the difference between muscles were small and often not significant (P>0.05).

Meat packed in CO_2 for 30 days had higher vitamin E concentrations than meat packed in air for 5 days (P<0.001). The mean concentrations for CO_2 and air packed meat was 4.15 and 3.85 (LSD 5% = 0.24). The significance and reasons for this finding is unknown. The conclusion could be made that vitamin E is protected by CO_2 packaging. However the assay used to measure vitamin E concentration does not discriminate between reduced and oxidised forms so the activity of the vitamin E in the different packagings is unclear.

The concentrations of vitamin E in muscles of lambs grazing green pasture were similar to the levels found in lambs fed a pellet ration with 150ppm of vitamin E added.

5.4.2. Practical considerations for supplementing vitamin E

Results from experiment 2 demonstrated the relationship between the level of inclusion of vitamin E in a feed ration and the time taken for muscle vitamin E concentration to reach the threshold level of 3 mg/kg. The lower the level of inclusion then the longer the feeding period needs to be for repletion to occur (Figure 9). This relationship can be used as the basis for advice to farmers about vitamin E inclusion rates. The rate of repletion was slower in the ST than the SM (Figure 9, 10). However the value of vitamin E is greater for the SM than the ST, so recommendations based on the SM will be sufficient for the whole carcass. The results suggest that a minimum feeding period of 2 weeks is required if an inclusion rate of 275ppm is used. If the feeding period is for 1 week then an inclusion rate of 400ppm is required.

A level of 275ppm is greater than the recommended level for lamb health requirements (Fry *et al.*, 1996), and therefore the extra cost associated with this may be of interest to producers. For an inclusion rate of 275ppm the cost of vitamin E for a 14 day feeding period would be 1cent/kg carcass weight, or 20cents per lamb (Figure 11)



Figure 9 Estimated cost of vitamin E feed supplement



Figure 10 The change in vitamin E concentration in *m. semimembranosus* with time





5.4.3. Animal production implications for vitamin E supplementation

There was no effect (P>0.05) of vitamin E supplementation on liveweight in experiment 2 (Figure 12)

Figure 12 The change in liveweight with time (experiment 2)



5.5. Interactions between treatments

Some important interactions occurred that made interpretation of the data complex. An example of this is the interaction between electrical stimulation, ageing and vitamin E treatments that occurred for SM in experiment 2. Whilst there were no main effects of electrical stimulation in the SM there was a significant 3 way interaction (P<0.05). Electrical stimulation reduced colour stability for 5 day air aged meat particularly when vitamin E was low (Figure 13). Strategies that improve colour stability could involve changes along the supply chain that include lamb production, processing and packaging elements.



Figure 13 The interaction between ageing electrical stimulation and predicted shelf life of the SM

□ Day 5 □ Day 10 □ Day 20 □ Day 30

5.6. Industry application of colour stability measurement

5.6.1. Consumer perception of brownness

Purchasing choices made by consumers surveyed at the Captain Stirling IGA store were clearly influenced by the colour of meat in relation to brownness.

	Percentage (%) of people			ole
	Agree	Indifferent	Disagree	Don't know
Brown meat is ok to eat	49	12	30	9
(if not past its use-by-date)				
Red meat is an important part of my diet	93	6	1	0
I would eat brown meat (if not past its use-by-date)	48	10	41	1
Colour influences which piece of meat I select	84	7	7	2

Table 5 The importance of meat colour to participants buying decisions

The results from this survey suggest that there is a threshold value for oxy/met ratio at which consumers distinguish between red and brown colour (Figure 14). That being the case

Figure 14 The relationship between oxy/met ratio and consumer score



Colour score (y) can be predicted from oxy/met ratio (x) using the equation:

y=29.22 + 38.65/[1 + e^{-5.38(x-2.9234)}]

R² = 0.395 (F_{3, 495} = 190.37; p<0.001))

5.6.2. Differences between Hunterlab reflectometers

The translucency of meat influences reflection of light from the surface and this was found to interact with the size of the reading frame in Hunterlab reflectometers. The 45/0L model was found to read higher levels for the wavelength of 630nm compared to the 45/0S. The magnitude of this difference depended on the colour of the meat (Figure 17) and was greatest when myoglobin concentration was low.



Figure 15 Light reflectance at 630nm for the Hunterlab 45O/S and 45O/L

The difference in light reflectance measurement at this wavelength caused a large effect on oxy/met ratio and the measurement of colour change during shelf display (Figure 15, 16) For this reason the type of Hunterlab spectrophotometer needs to be specified when reporting data. For ease of comparison we recommend that one type of Hunterlab be used and that the 450/L will be yield more meaningful results than the 450/S for the purpose of measuring colour stability of lamb meat.

Figure 16 Comparison of the change of the oxy/met ratio in time for lamb loin between obtained with 45O/S and 45O/L Hunterlab spectrophotometers



5.6.3. Description of colour change

Colour stability is a complex measurement because it involves a number of colour measurements over time. To simplify colour stability data to allow communication to industry people, some investigation into the different ways of describing colour stability were undertaken. Different ways of expressing colour change include:

1. Rate of change of oxy/met over display time

The non linearity of the relationship between colour and time makes statistical analyses of colour stability data difficult. The nature of the relationship depends on the muscle. Two different statistical methods used were: to fit a polynomial function to the data and to use the smoothing splines method to fit a curve. The smoothing splines method allows closer fit of the model to the data when the data does not follow a known function but is more difficult to do.

2. Time to reach a benchmark oxy/met ratio

The results from the consumer survey (experiment 3) suggest that an oxy/met ratio of 3.5 is the level at which consumer acceptance of meat changes. However the time to reach a set oxy/met ratio benchmark is a function of the starting value as well as

the rate of change with time. Metmyoglobin production occurs at the surface after cutting so a change in the starting value is unlikely to be due to metmyoglobin formation. Treatments can affect both the starting value and the rate of change of oxy/met ratio and these effects may be accounted for by different mechanistic reasons.

An example of this is the comparison between treatments from experiment 1. When packed in air for 5 days (Figure 17), there was a clear difference in the time required for the SM to reach the benchmark value of 3.5 between treatments. This was reached after 30h, 40h, and 60h approximately for green pasture, dry feed and feedlot supplemented with vitamin at 150ppm respectively. The difference between the dry feed and the feedlot ration was due to the different slopes of the curve hence the rate of metmyoglobin production. The difference between the feedlot ration and the green pasture was due to differences in the starting values.

When packed in CO_2 for 30 days (Figure 18), the benchmark value of 3.5 was reached after 20h, 30h, and 50h approximately for dry feed, green pasture, and feedlot supplemented with vitamin at 150ppm respectively. In this case the slopes of the curves were the same for each of the treatments, so this difference was due to the starting value rather than the change in rate of colour change.

3. Oxy/met ratio at a standard display time.

The industry standard currently is 48 h and the goal of retailers is to extend this to 60h so that less meat has to be discounted prior to sale. This method effectively is the inverse of the time to reach a benchmark value for oxy/met ration so the same comments apply to both methods.

The value of using benchmark is that they allow simple comparisons between industry practices and to value interventions financially in a commercial context.

For industry applications the time to reach a benchmark or the oxy/met ratio at a standard time may be sufficient information and be more useful than a detail description of the colour relationship with time. In a research context both the slope and the starting value that describe any relationship between oxy/met ratio and time are likely to be important as different mechanisms may be involved. In fact further work is required to understand the difference mechanisms that change the slope and the starting values respectively.



Figure 17 The change in oxy/met ratio over time for air packed SM aged for 5 days

Figure 18The change in oxy/met ratio over time for CO₂ packed SM aged for 30 days



6. Success in achieving objectives

Objective	Specific Aims	Outcome	
	Medium voltage electrical stimulation	Achieved	
Description of the effects of new processing technologies on colour stability	CO ₂ packaging	• Partly achieved, subsequent work planned to compare CO2 packaging with vacuum packaging at different ages	
	Vitamin E supplementation	 Rates and timing for feed supplementation achieved 	
Extend shelf life beyond 48 h	Data synthesis	Document ready for industry consultation	
Industry applicable method to describe colour stability		 Nature of oxy/met time relationship described Comparison of Hunterlab spectrophotometers 	
	Benchmark of consumer acceptance	 Categorised cuts Pilot survey completed further work required 	

Table 6 Success in achieving objectives

7. Impact on Meat and Livestock Industry – now and in five years time

7.1. Industry Impact now

This study has confirmed that new processing technologies adopted by the Woolworths supply change will not be detrimental to lamb meat colour stability. This underpins the eating quality improvements gained by electrical stimulation and ageing.

7.2. Extension undertaken during project

7.2.1. Presentations

- Another reason to use Vitamin E; Sheep updates Narrogin 2006
- Project update Woolworths/ V&V Walsh management April 2007

7.2.2. Published Articles

- Lamb meat colour- a producer, processor and retailer issue. Ovine Observer June 2006
- Does vitamin E improve growth rate? Ovine Observer September 2006
- Vitamin E as a dietary supplement improves sheep meat colour stability. Sheep CRC conference Orange 2006
- Vitamin E supplement keeps lamb meat red longer. Australian Farm Journal May 2007

7.2.3. Sheep CRC Factsheets

• Quality sheep meat: Meat colour and shelf life

7.3. Industry impact in 5 years

This project has provided tools to improve lamb meat colour stability in the future. Further consultation with industry partners is required to determine how this information could be commercialized.

8. Conclusions and Recommendations

8.1. Recommendations

- Packaging meat as primal cuts in carbon dioxide for 10 days produces a highly acceptable colour and colour stability for lamb meat.
- Medium voltage electrical stimulation can be used without detrimental effects on retail display time under the current 48h scenario.
- Lamb meat should not be aged for longer than 10 days unless the vitamin E status of the lambs prior to slaughter can be assured. Extended ageing to 30 days will reduce colour stability to an unacceptable level when the vitamin E concentration in meat is low.
- Supplementation of the lamb finishing diet with vitamin E can improve the colour stability of lamb meat. For lambs backgrounded on dry pasture, the recommended rate of inclusion of vitamin E in a feedlot diet is 250ppm. This should be fed for a minimum period of 2 weeks prior to slaughter.
- To ensure that meat is attractive to consumers, the oxy/met ratio of lamb meat should not fall below 3.5 during the retail display period.
- Display period could be extended from 48h to 60h but would involve consideration of cut, lamb history, and ageing time.

8.2. Recommendations for further research

- The effects of green pasture on lamb meat colour and colour stability
- Alternative methods of administering vitamin E
- Consumer perception of colour change in different lamb meat cuts
- The influence of other antioxidants and pro-oxidants on colour stability
- Seasonality of colour stability
- Economic evaluation of colour stability in Australia

9. Appendices

9.1. Appendix 1 Consumer survey questionnaire

By completing this questionnaire you will help us determine consumer sensitivity to changes in red meat colour or the colour of red meat. None of your answers will be used individually or given to anyone else. Please do not hesitate to place comments beside any questions.

1. On average, how often do you eat beef? (please circle)

daily more than 2 times a week 1-2 times a week fortnightly monthly never

2. On average, how often do you eat lamb? (please circle)

daily more than 2 times a week 1-2 times a week fortnightly monthly never

Background questions

Your answers to these questions will be helpful for us to check that the sample of people used is representative of the population as a whole.

3. What is your post code? _____

- 3. Please circle your age group
- 4.

<20 20-29 30-39 40-49 50-59 60-69 70+

5. Please circle your gender?

Male

Female

6. Please circle the highest level of education you have completed?

Primary	y School	High School	TAFE	University	Other
	/	0		<u> </u>	

If you *do not* eat meat, please place your questionnaire in the box and we thank you for your time.

If you *do* eat meat please turn over the page and take part in the following activity.

Look at each sample of beef and lamb provided and place a vertical line at the point on the colour scale that you think is right. Also please tick the last box if you would be happy to eat this sample of meat or a cross if you would not like to eat it.

 \checkmark

Colour EXAMPLE Red-----Brown

Comple	Calaur		\checkmark = would eat
Sample	Colour		✗ = would not eat it
1	RedI	Brown	
2	Red	Brown	
3	Red	Brown	
4	RedI	Brown	
5	RedI	Brown	
6	RedI	Brown	
7	RedI	Brown	
8	RedI	Brown	

7. For **BEEF**, look at samples 1- 8 and fill in the table below.

For beef, put a vertical line at the point on the scale where you think the colour represents the best quality for eating.

Red-----Brown

				✓ = would eat		
Sample	Colour		× =	would	not	
			eat it			
9	RedI	Brown				
10	RedI	Brown				
11	RedI	Brown				
12	Red	Brown				
13	RedI	Brown				
14	Red	Brown				
15	Red	Brown				
16	Red	Brown				

9. For LAMB, please look at samples 9-16 and fill in the table below .

For lamb, put a vertical line at the point on the scale where you think the colour represents the best quality for eating.

Red-----Brown

Attitudes Towards Red Meat Colour

11. When selecting the meat that you eat do you base your choice on colour? (please circle)

YES NO

Please tick the box that best reflects your view for each of these statements.

	Strongly Agree	Agree	Indifferent	Disagree	Strongly Disagree	Don't Know
Meat that is light red in colour is always of better						
Brown meat that is not past its use-by- date is ok to eat						
I think dark red meat is best						
I consider price to be the most important factor						
Red meat is an important part of my diet						
The colour of fresh raw meat is irrelevant once						
I prefer not to eat light red meat						
I would eat brown meat that is not past its use-by-						
Colour influences which piece of meat I select						

13. If you have any comments that you would like to make regarding this topic, please write them in the space below.

9.2. Appendix 2 Consumer Benchmarks commercialising lamb meat colour research findings summary

9.2.1. Background

For research purposes meat colour stability is measured by the reflection of light at 2 wavelengths (580 nm and 630nm) over time. Reflection of these different wavelengths of light from meat changes with shelf display time as oxymyoglobin (red pigment) becomes oxidised to metmyoglobin (brown pigment).

9.2.2. Colour perception

To be meaningful in a commercial context, objective measurement of light reflectance obviously needs to correlate with consumer perceptions of colour. Preliminary research suggests that this is the case. In fact consumers tend to perceive that meat colour changes from red to brown discretely at a threshold level rather than as a continuous change from one extreme to the other.

9.2.3. Colour Benchmarks

This finding is the basis of using a benchmark for comparing the success of various treatments designed to improve colour stability. Consumers perceive meat to be red in colour when the ratio is above the threshold value and brown when the ratio is below the threshold value. However the translucency of meat is a complicating factor such that the nominal value of this benchmark depends on the optical geometry of the spectrophotometer used to measure light reflectance. A value of 3.5 appears to be the benchmark for the threshold between red and brown for a Hunterlab 45/L0 spectrophotometer.



Figure 19 Correlation between consumer perception of colour and light reflectance

9.2.4. Consumer attitudes

The importance of meat colour to consumers as a basis for meat purchase decisions is demonstrated in table 1.

	Percentage (%) of people			
_	Agree	Indifferent	Disagree	Don't know
Brown meat is ok to eat*	49	12	30	9
Red meat is an important part of my diet	93	6	1	0
I would eat brown meat *	48	10	41	1
Colour influences which piece of meat I select	84	7	7	2

Table 7 The attitudes of consumers to meat colour

*providing it is not past its use-by-date

9.2.5. Colour benchmarks

Colour stability is a function of both colour and time. However supermarkets tend to make decisions using time alone and discount meat when a standard time period has elapsed.

Meat should therefore have a reflectance ratio greater than the benchmark figure (3.5) at the time discounts are applied (48 hours) otherwise consumers will perceive

the meat available for purchase to be brown in colour and their purchasing decisions will be affected accordingly.

9.2.6. The current situation

In store data

Currently we have no data collected from "in store" for the time lamb meat stays on the shelf or the percentage that has a colour value above the colour benchmark (3.5) until the time of purchase.

Hence we cannot make any objective assessment about the success of the current practice of a 48 hour shelf life.

Management factors

We do have evidence that lamb production factors including lamb age, genotype and nutrition influence lamb meat colour and that these factors change over a 12 month production cycle. We also know that the shelf life of lamb meat can be extended to 60 hours provided certain conditions in relation to these management factors are met.

9.2.7. The importance of cut

Research has shown that different muscles vary greatly in the rate at which they discolour during shelf display. Generally the more pigmented a muscle is then the more oxidative and unstable in colour it will be during shelf display.

Classification	Expected change (%)*	Muscle	Commercial Cut
Colour Stable	< 5	m. semitendinosus	Eye of Silverside
		m. biceps femoris	Silverside
Colour	0-25	m. rectus femoris	Knuckle
Intermediate		m. longissimus thoracis et	Loin
		m. triceps brachii	Shoulder
Colour		m. semimembranosus	Topside
Unstable	> 25	m. gluteus medialis	Rump
		m. psoas major	Scotch fillet

Classification of different cuts for colour stability

* This is the expected amount of change in reflectance ratio (630/580) for meat aged 5 days post slaughter then sliced and displayed for 48 hours.

9.2.8. Linking supply chain management

Meat colour can be influenced by management during lamb production, lamb processing and retail.

9.2.9. Production on farm

Lamb carcass specifications for weight and fat score stay constant throughout the year. However until about December lamb supply consists mainly of sucker lambs that are 3-5months of age, predominantly crossbred genotype that have not been weaned. After December lamb supply changes to carry over lambs; 8-12 months of age, merino genotypes as well as crossbreds, that have been weaned.

9.2.10. Lamb age

Carry over lambs are older so their meat tends to have more pigment and be darker in colour than for sucker lambs. Meat from carry over lambs may also be more variable in colour than meat from sucker lambs.

9.2.11. Genotype

Meat from Merino lambs may be more unstable in colour than meat from crossbred lambs. Extended shelf life probably cannot be recommended for merino meat with current understandings.

9.2.12. Vitamin E

Vitamin E is important particularly for meat that has been aged prior to display. If meat is to be aged for longer than 5 days then the vitamin E concentration should be greater than 3 mg/kg for good colour stability. Otherwise meat may become brown in colour within a 48 hour display period.

Supplementation with vitamin E improves colour stability of all muscle types although the size of this effect is dependent on the muscle. Colour unstable muscles benefit more from vitamin E than do colour stable muscles. It is important to realise that vitamin E supplementation does not change the differences between different muscle types for colour stability. In fact colour stable muscles often have lower vitamin E concentrations naturally than do colour unstable muscles. Other benefits of vitamin E are to make meat lighter in colour and more tender.

Lambs gain vitamin E from green feed so if they have had access to dry feed only for greater than 6 weeks they will likely be vitamin E deficient. Vitamin E muscle concentrations can be boosted to sufficient levels for colour stability by supplementing lamb finishing diets with synthetic vitamin E at the rate of 250mg/kg for 2 weeks prior to consignment for slaughter.

9.2.13. Processing

Electrical stimulation

Medium voltage stimulation has no detrimental effect on colour stability but makes meat appear lighter in colour.

Gas packaging

Packaging primal cuts in carbon dioxide makes meat more red in colour particularly at the commencement of the display period.

Ageing

Colour change doesn't occur until after the meat has been sliced for display. However peroxidation of fats during ageing speeds up this process once the meat has been sliced. Ageing reduces colour stability of all muscle types including those classified as colour stable for a 5 day ageing period. Generally the longer the ageing period the greater the less colour stable meat will be.

9.2.14. Commercialising improvements to shelf life

The research to date has benchmarked colour stability in a range of cuts. Commercial gain can be made either by reducing cost or increasing product value.

9.2.15. Cost reduction

Increasing the shelf life from 48 hours to 60 hours would reduce the quantity of meat discounted hence the cost of retailing meat. Our results suggest that even colour unstable cuts could be displayed for 60 hours under certain management scenarios.

9.2.16. Supply consistency

Adoption of these research findings could improve the consistency of quality on a year round supply basis from different production systems.

9.2.17. Recommendations for extending shelf life to 60 hours

Table 3 has been synthesised from different experimental results from our research work and is a starting point for working out a strategy to extend the shelf life of lamb meat.

These guidelines could be recommended only after more development work is done:

- 1. At the store level with consumers and commercial product.
- 2. At the supply chain level to link sheep and carcass management strategies.

Finish	Vitamin E status	Ageing period (d)	Cut colour type			
1 111311		Ageing period (d)	Stable	Intermediate	Unstable	
		5	۲	•	•	
Feedlot	High	10	۲	•	۲	
	(250 ppm)	20	۲	•	۲	
		30	х	•	۲	
		5	х		x	
Green Pasture	Medium (150ppm)	10	۲	9	x	
		20	۲	9	x	
		30	Х	x	x	
	Low	5		9	Ð	
Feedlot	(30 ppm)	10		9	x	
		20	€	•	x	
		30	х	х	x	
	Depleted	5	х	х	x	
Dry pasturo	(0ppm)	10	х	х	x	
biy pasture		20	х	х	х	
		30	Х	x	х	

Extended shelf life (60h) recommendations for crossbred lamb meat

X = 60 hours is not recommended for this cut by management combination

• = 60 hours is recommended for this cut by management combination

For example 60 hours is not possible in any scenario if lambs are finished on dry pasture. However 60 hours is possible in all cuts for lambs finished in a feedlot with a high vitamin E diet and the meat is aged for less than 20 days.

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