

# final report

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## **Impact of CT scan radiation on colour stability in lamb & beef**

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## Abstract

This study investigated the effect of Computerised Tomography imaging (CT scan) on the oxy/metmyoglobin ratio, hue and  $L^*a^*b^*$  scores of *m. longissimus dorsi* from both beef and lamb. Beef and lamb *m. longissimus dorsi* were divided into 4 portions and randomly allocated into one of the following treatments; CT 30 day aged; CT fresh; control 30 day aged; control fresh. After the designated ageing period, samples were cut and displayed under retail conditions, with colour measurements taken over a 96 hour period. CT scan had little effect on the colour of both lamb and beef across all colour parameters measured. There were small effects observed in CT aged samples for ratio, hue,  $a^*$  and  $b^*$  values, however it is unlikely that the commercial shelf-life of the product would be affected. Other factors such as ageing, species and vitamin E concentration were found to play a much greater role in the colour stability of both beef and lamb than the CT scan treatment. Aged *m. longissimus dorsi* clearly had a worse colour stability than the fresh packaged samples, while beef was a lot more colour stable than lamb. It appears that CT scan for the purpose of body composition determination will not have any commercially relevant impact on colour stability of both beef and lamb.

## Executive Summary

- Computerised Tomography imaging (CT scan) is currently being investigated as a means of providing skeletal coordinates for automated boning plants as well as providing on-line yield prediction. CT scanners produce a small amount of radiation output, and given other evidence that irradiation of meat (for sterilisation purposes) reduces colour stability of fresh product, this was raised as a potential concern.
- Thus a study was carried out to investigate the effect of Computerised Tomography imaging (CT scan) on the oxy/metmyoglobin ratio, hue and L\*a\*b\* scores of *m. longissimus dorsi* from both beef and lamb.
- Beef and lamb *m. longissimus dorsi* were exposed to the following treatments; CT 30 day aged; CT fresh; control 30 day aged; control fresh.
- Over a 96 hour period CT scan had little effect on the colour of either lamb or beef across for any of the colour parameters measured. There were small effects observed in CT aged samples for ratio, hue, a\* and b\* values, however it is unlikely that the commercial shelf-life of the product would be affected.
- It appears that CT scan for the purpose of body composition determination will not have any commercially relevant impact on colour stability of both beef and lamb.
- Given these findings, further work into the development of a lamb or beef CT scan system should not put at risk the shelf life of these meats.

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# 1 Background

Surface meat colour is an important trait that consumers consider when purchasing a cut of meat. If the surface appears discoloured and brown the consumer will most likely choose not to buy that cut, causing a massive economic loss to the Australian meat industry. The surface colour of meat is due to the chemical state of the muscle pigment myoglobin (Fox 1966). The red pigment, oxymyoglobin, can be oxidized to form the brown metmyoglobin. The surface of meat will appear brown when the ratio of oxymyoglobin to metmyoglobin becomes too low (Hunt 1980).

A variety of factors can influence the colour stability of meat products. These factors can range from the animals diet to how the meat is packaged in the retail stores. To optimise the colour stability, these factors are managed to keep the amount of oxidation. Ionizing radiation (irradiation) has been used in the food industry for many years, preserving foods by killing microbes (Brewer 2004). Irradiation however, can produce damaging and highly oxidative free radicals such as the hydroxyl radical which can oxidize myoglobin directly, or indirectly through the oxidation of lipids (Thakur and Singh 1994).

Computerised Tomography (CT) imaging, also known as Computer Aided Tomography, is a form of low level irradiation. CT has the capacity to differentiate tissue types in live animals and has a high level of accuracy in predicting body composition (Afonso 1992; Jones *et al.* 2002; Jopson *et al.* 1995; Lambe *et al.* 2003; Young *et al.* 1996). Determining carcass composition using CT has also shown strong correlation with the traditional dissection methods (Afonso 1992; Lambe *et al.* 2003). By incorporating this technology, producers are able to meet the demand for larger leaner carcasses. In addition to this, the newest CT scanners are able to scan entire carcasses at abattoir chain speed which makes them suitable for carcass grading and yield prediction. However, it has not yet been

established whether the low level of radiation output from the CT scanner will affect colour stability of fresh meat.

## 2 Project Objectives

The purpose of this present study was to test the hypothesis that CT scanning carcasses for body composition will reduce the colour stability and shelf life of lamb and beef.

## 3 Methodology

### 3.1 Animals and Treatments

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14 lamb loins and 15 beef cube rolls (lumbar region of *m. longissimus dorsi*) were purchased from a commercial abattoir. Lambs were fed on a dry annual pasture and carcasses had an average weight of  $21.7 \pm 0.49$  kg and a fat score of 3. Beef were sourced from a feedlot, and the live weight ranged between 400-450 kg. Each muscle was divided into 4 equal proportions and was randomly allocated to one of the following treatment groups; CT aged; CT fresh; control aged; control fresh. Control groups were those that were not CT scanned. Aged groups were packaged in vacuum packs for 30 days before cutting for retail display, while the fresh groups were packaged loosely in air for 5 days before cutting for retail display.

### 3.2 Colour Measurements

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Colour measurements were made using a Hunter Lab Mini Scan XE Plus (model No. 45/0-L, Hunter Associates Laboratory Inc., Reston VA, USA), Using C set as the light source with a aperture set to 10. Readings were taken every 12 hours after the meat had been sliced (2cm thickness), placed on black Styrofoam trays and over wrapped in polyvinyl chloride wrap. The meat was stored at 4°C in a

fridge fitted with fluorescent lights. Colour was measured using  $L^*a^*b^*$  scores ( $L$ = lightness darkness,  $a^*$  red to green,  $b^*$  yellow to blue), and hue angle was calculated as in equation 1.

$$\text{Hue} = -\tan(a/b)$$

**equation 1**

Myoglobin oxidation was predicted from the oxy/metmyoglobin ratio calculated from the ratio of light reflectance at 630 and 580 nm . A ratio score of 3.5 was used as a consumer discrimination point, where the browning of meat becomes evident.

### **3.3 Vitamin E Analysis**

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The vitamin E content of muscle was measured using high performance liquid chromatography with fluorescence detection (McMurray and Blanchflower 1979). Tissues were saponified by the method of Bieri *et al.*(1961), before extraction with hexane.

### **3.4 CT Scanning**

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Meat was scanned using a Picker PQ 5000 spiral CT scanner located at the Murdoch University Veterinary School. Settings used for the scan were: pilot scan length of 512mm, position was prone, field of view set at 480, Index 20, slice thickness 10 mm, mA 150, revs 40, pitch 1.5 and algorithm standard.

### **3.5 Statistical Analysis**

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The effect of CT scanning, aging, species and time on the ratio of light reflectance at 630 and 580 nm was determined using a linear mixed effect model, where CT scanning, aging treatment, and species were used as fixed effects, and time was used as a covariate in both the linear and curvilinear forms, using SAS (SAS Institute 2001). Animal within species was the random term.

Interactions between all of the fixed effect and covariate terms were tested and removed ( $P>0.05$ ) if not significant. Similar models were used to analyse Hue,  $a^*$ , and  $b^*$  values.

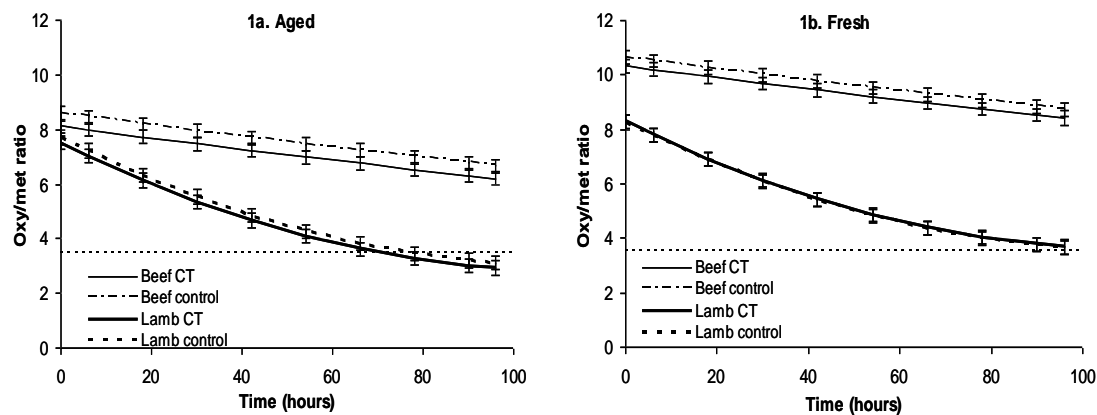
## 4 Results and Discussion

### 4.1 Results

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#### 4.1.1 Oxy/metmyoglobin ratio

In both fresh and aged beef CT scanning had a small negative effect ( $P<0.05$ ) on the ratio of light reflectance at 630 and 580 nm, causing an initial reduction of about 0.3-0.5 ratio units at the first reading, which was maintained throughout the rest of the retail display period (figure 1a;b). This difference in ratio did not change over time, thus CT scanning did not affect colour stability in beef. There was no effect of CT scanning in either aged or fresh lamb meat.



**Figure 1** The effect of CT scan on the oxy/metmyoglobin ratio over 96 hours retail display in a) aged beef and lamb *m. longissimus dorsi*; and b) fresh beef and lamb *m. longissimus dorsi*.

The ratio of light reflectance at 630 and 580 nm declined more rapidly ( $P<0.05$ ) over the retail display period in lamb meat compared to beef (figure 1a;b), indicating a faster rate of metmyoglobin



accumulation in lamb. Both aged lamb and beef had a more rapid decline in ratio than the fresh treatment groups ( $P < 0.05$ ). Aged lamb reached an unsatisfactory ratio of 3.5 after 60 hours retail display while all other treatment groups maintained a satisfactory colour over the entire display period.

All statistical interactions can be observed in Table 1 below.

**Table 1.** F-values for the effect of CT scanning, aging, species and time on the ratio, hue, a\*, and b\* values of lamb and beef *m. longissimus dorsi* muscle.

Effect	Ratio		Hue		a*		b*	
	NDF;DDF	F val	NDF;DDF	F val	NDF;DDF	F val	NDF;DDF	F val
Time	1;1123	268.51***	1;1119	16.29***	1;1113	12.49***	1;1121	49.61***
Time2	1;1123	36.93***	1;1119	15.95***	1;1113	21.19***	1;1121	0.16
Species	1;27	18.12***	1;27	32.11***	1;27	35.97***	1;27	11.62***
CT	1;1123	17.89***	1;1119	0.57	1;1113	0.01	1;1121	1.51
Age	1;1123	638.93***	1;1119	50.44***	1;1113	0.5	1;1121	84.39***
Time x Species	1;1123	91.06***	1;1119	135.97***	1;1113	160.87***	1;1121	29.39***
Time x CT	-	-	1;1119	4.13**	1;1113	5.64**	-	-
Time x Age	-	-	1;1119	14.79***	1;1113	6.14**	1;1121	26.28***
Species x CT	1;1123	7.54***	1;1119	7.16***	1;1113	4.16**	1;1121	14.61***
Age x Species	1;1123	176.38***	1;1119	3.84*	1;1113	37.27***	1;1121	6.52**
Age x CT	-	-	1;1119	1.58	1;1113	2.02	-	-
Time x Age x CT	-	-	-	-	1;1113	5.37**	-	-
Time x Species x CT	-	-	-	-	1;1113	1.01	-	-
Time x Age x Species	-	-	-	-	1;1113	19.87***	-	-
Age x Species x CT	-	-	1;1119	8.02***	1;1113	4.65**	-	-
Time2 x Species	1;1123	29.06***	-	-	1;1113	38.4***	1;1121	6.31**
Time2 x Age	-	-	1;1119	9.58***	1;1113	0	1;1121	6.97***
Time2 x Age x Species	-	-	-	-	1;1113	14.09***	-	-
Time x Age x Species x CT	-	-	-	-	1;1113	8.2***	-	-

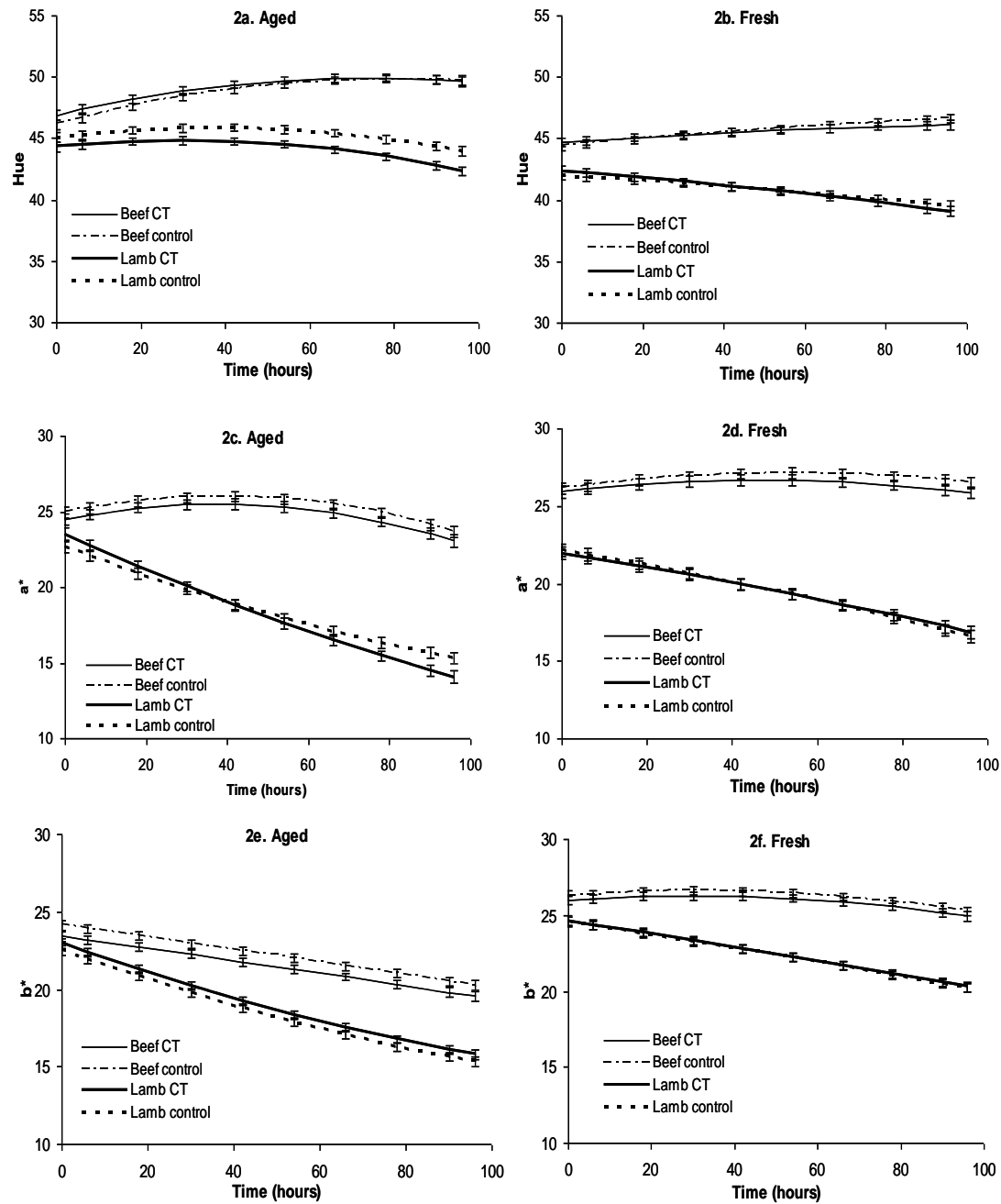
Level of significance denoted by \*\*\* P< 0.01; \*\* P< 0.05; \* P< 0.1. NDF, numerator degrees of freedom; DDF, denominator degrees of freedom.

#### 4.1.2 Colour Parameters

The hue of lamb decreased (more red) while the beef slightly increased (less red) over time of display. Meat from aged CT lambs had a lower hue angle and thus was redder than the controls. CT scan treatments had a slightly steeper decline in hue angle over the display period when compared to that of the controls (figure 2a;b).

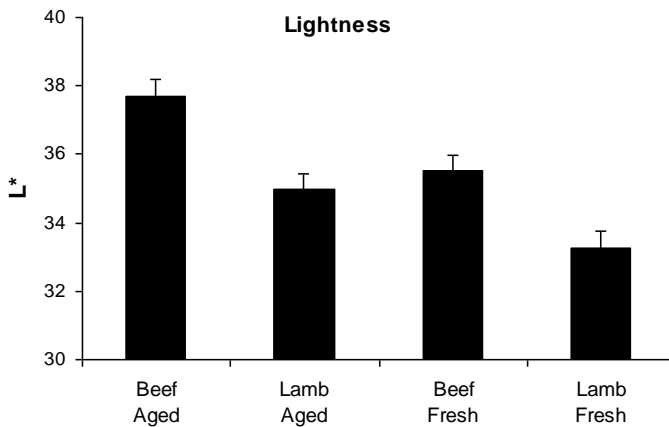
CT scanning marginally decreased ( $P<0.05$ ) the  $a^*$  value in the aged lamb when compared to the control (figure 2c;d), but only after about 70 hours of retail display. There was no effect in the fresh lamb treatment, or in either of the beef treatments. The  $a^*$  value in lamb decreased faster than beef ( $P<0.05$ ), thus lamb was less red than beef. This rate of decline was faster ( $P<0.05$ ) when the meat was aged.

The  $b^*$  value decreased with time, with this decrease more rapid in lamb ( $P<0.05$ ) compared to beef (figure 2e;f). The  $b^*$  value also decreased more rapidly for the aged meat compared to the fresh, in both lamb and beef. There was no effect of CT scan on the  $b^*$  value with time.



**Figure 2.** The effect of CT scan on hue (a;b),  $a^*$  (c;d) and  $b^*$  (e;f) values over 96 hours retail display in aged and fresh beef and lamb *m. longissimus dorsi*.

CT scanning had no effect on the L\* value. Lamb was darker than beef ( $P<0.05$ ) and aged meat was lighter ( $P<0.05$ ) when compared to the fresh product (figure 3).



**Figure 3.** The effect of aging for 30 days on the lightness (L\*) of beef and lamb *m. longissimus dorsi*.

#### 4.1.3 Muscle Vitamin E

The average vitamin E concentrations of the Beef LD muscle were  $2.40 \pm 0.078$  mg/kg tissue ( $\pm$ sem), while the same muscle in lamb had much lower concentrations ( $P<0.05$ ) with  $0.64 \pm 0.036$  mg/kg tissue ( $\pm$ sem).

## 4.2 Discussion

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CT scan had little effect on the colour of both lamb and beef across all colour parameters. CT Lamb reached an unacceptable oxy/metmyoglobin ratio of 3.5 (known to be the point at which consumers visually discriminate against this product based on colour) at the same time as the controls in both the fresh and aged meat, therefore there was no increase in metmyoglobin formation due to CT

scan. CT beef did have a slightly higher proportion of metmyoglobin over the retail display period, particularly for the aged product. However, this effect was small and the oxy/metmyoglobin ratio was still well above the consumer discrimination threshold of 3.5 after 96 hours retail display, thus would not have had any negative effect on the shelf life of these products commercially.

In contrast to this study other authors have shown the effects of irradiation maybe detrimental to the colour stability of red meat products (Kim *et al.* 2002; Millar *et al.* 2000; Murano *et al.* 1998; Nanke *et al.* 1998) . This finding is not surprising when considering the difference between radiation dosages. A typical CT scan for human medical purposes provides about 30 mGy of radiation, where as in previous studies the doses have ranged from 1-10 kGy , a  $10^6$  fold increase in radiation. Additionally, the *m. longissimus dorsi* is a relatively colour stable muscle (Hood 1980; O'Keefe and Hood 1982) and this may also be responsible for the little effect observed. It is also important to note that irradiation of oxymyoglobin often results in metmyoglobin, while irradiation of metmyoglobin may result in the formation of a red product (Ginger *et al.* 1955), which can explain the improvement of colour often observed in the presence of irradiation (Brewer 2004; Millar *et al.* 1996).

Nanke *et al.*(1998) found that irradiated vacuum packaged beef was browner than controls, and had lower  $a^*$  values and increased  $b^*$  values. This contrasts with the present study where we found the  $b^*$  value (yellowness) to decrease for both beef and lamb, which contradicts comments made by Brewer (2004) that irradiation increases yellowness across all species. The  $L^*$  value (lightness) in both species increased with aging, but there was no further increase with exposure to irradiation, as suggested by Nam and Ahn (2003).

Although significant effects of CT scan were observed for ratio, hue,  $a^*$  and  $b^*$  values (table 1) in the aged samples, these effects were only marginal and it is unlikely that they would effect the commercial shelf-life of the product. There was very little variation in the slopes, or rate of decline of each colour parameter due to CT scan in the aged product, thus the small difference evident in aged

CT beef is due to differences in the starting values. The rate of oxygen consumption will decrease with aging, known to cause variation in oxygenation layers in meat (O'Keefe and Hood 1982). This and the possible interaction of oxidative stress added by CT scan may be mechanistically responsible for this CT scan aged effect.

It has been shown irradiation will change the fatty acid composition in beef when exposed at the kGy level (Brito *et al.* 2002; Chen *et al.* 2007; Yilmaz and Gecgel 2007). In these cases the level of polyunsaturated fatty acids declined while the trans-fatty acids increased with level of irradiation. There has also been an observed increase in the formation of thiobarbituric acid reactive substances (an indicator of lipid peroxidation) when beef was exposed to these levels of irradiation (Chen *et al.* 2007; Formanek *et al.* 2003). Evidence shows that the total amounts of polyunsaturated fatty acids were decreasing due to irradiation. The double bonds in these fatty acids were most likely being altered, resulting in the formation of free radicals and other oxidative products. This mechanism should account for the effect of CT scan observed in the aged samples, where during the aging period the free radicals produced had greater chance to alter the chemical state of myoglobin.

A majority of the observed effect of CT scan was only in the beef samples. When considering the composition of lipids in beef is much more unsaturated than that of lamb (Droulez *et al.* 2006), irradiation is likely to have a more marked effect on the lipids in beef, in turn having a greater effect on the formation of metmyoglobin.

An important finding in the present study was the difference in colour stabilities between lamb and beef *m. longissimus dorsi*. It was found that beef clearly had a much better colour stability based on all the colour parameters measured. However, this contradicts Gutzke *et al.* (1997) who reported that the colour stability of meat from sheep was no worse than that of beef. The beef used in the present study had a 4 fold higher muscle vitamin E concentration than that of lamb, which were close to deficient levels. Vitamin E has been shown to improve the colour stability of both beef (Arnold *et al.*

1993; Faustman *et al.* 1989) and lamb (Wulf *et al.* 1995). However, previous studies in lamb carried out in this laboratory have shown that the improvements in colour stability brought about by Vitamin E would only explain about 2/5 of the difference evident between species in the present study. All parameters measured in beef had high starting points and low rates of decline when compared to lamb. This could possibly be explained by difference in oxygen uptakes of post mortem muscles, as lamb is likely to take up more oxygen than beef (Atkinson and Follet 1973) as lamb is more oxidative. The different rates of oxygen uptake, cause different levels of oxygenation in the meat and reflect in the brightness of the surface colour. It is likely that there is more underlying physiological difference between the two species which is responsible for the difference in colour stabilities.

Another possible mechanism explaining the species difference could lie in the variation in iron and zinc concentrations. Both minerals play a major role in cellular oxidative processes. Iron in lamb can vary from 1.6 to 2.3 mg/100g of tissue (Hazell 1982; Lin *et al.* 1988; Rhee *et al.* 1993) while beef may vary between 2.2 to 2.4 mg/100g of tissue (Hazell 1982; Rhee *et al.* 1993). Furthermore, 77 percent of iron in beef was from soluble fractions (myoglobin, haemoglobin, ferritin) while in lamb only 67 percent was from this soluble fraction (Hazell 1982). The percentage of iron available as ferritin in lambs was 4 fold to that of beef (Hazell 1982). Beef also has higher concentrations of zinc (Hazell 1982; Rhee *et al.* 1993), yet the partitioning between soluble and insoluble fractions is identical (Hazell 1982). It seems that there is more free minerals in lamb available to take part in oxidative processes, and thus it is proposed that this will reflect negatively on the colour stability.

## **5 Success in Achieving Objectives**

The primary objective of this work was to establish whether CT scanning would affect colour stability of lamb and beef meat. We have successfully demonstrated that there appears to be no commercially relevant effect of CT scanning on colour stability of fresh or aged meat.



## **6 Impact on Meat and Livestock Industry – now & in five years time**

CT scanning technology is currently being developed for the processing sector of the red meat industry as a tool to provide spatial coordinates for automated boning plants, and to determine lean meat yield. This work can now proceed without concern that it will have an economically adverse impact on colour stability.

## **7 Conclusions and Recommendations**

It seems that there is very little effect of irradiation from CT scan on meat colour, contrasting with other irradiation studies which used far greater levels of irradiation. It is particularly evident that other factors such as ageing, species and vitamin E concentration play a much greater role in the colour stability than CT. Aged *m. longissimus dorsi* clearly had a worse colour stability than the fresh packaged samples, while beef was a lot more colour stable than lamb. The *m. longissimus dorsi* is a highly colour stable muscle (Hood 1980; O'Keefe and Hood 1982), and it is possible that less stable muscles may be more effected by the irradiation from CT scan. However, for the purpose of using CT commercially to scan carcasses for body composition, it appears that the level of irradiation involved will not have any commercially relevant impact on colour stability.

Developmental work of CT scanning technology should proceed without concern that it will adversely impact on colour stability.

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