VITAMIN E SUPPLEMENTATION IMPROVES RETAIL COLOUR LESS IN HIGH INTRAMUSCULAR FAT LONG-AGED LAMB MEAT

H.B. Calnan[1,2], R.H. Jacob[2], D.W. Pethick[1,2] and G.E. Gardner[1,2]

[1] Cooperative Research Centre for Sheep Industry Innovation, University of New England, Armidale, Australia 2351

[2] Division of Veterinary and Life Sciences, Murdoch University, Murdoch Food Engineering Department, Australia 6150

The rate of lamb browning under retail display increases with ageing which is a concern for chilled exported Australian lamb. We tested whether high intramuscular fat concentration (IMF), oxidative capacity and pH accelerated the rate of meat browning, and whether Vitamin E supplementation could negate these effects, particularly in long-aged meat. 132 lambs were fed a low or high (30mg/kg or 275mg/kg) Vitamin E ration for 8 weeks prior to slaughter. Samples were taken 24 hours post-mortem from the *m.longissimus* to determine Vitamin E concentration, IMF, isocitrate dehydrogenase activity (ICDH) and pH. Additional samples were aged for 5, 35 and 70 days before being re-sliced, wrapped and placed under simulated retail display for 72 hours. Surface redness (reflectance at 630/580nm) was measured daily and analysed in a linear mixed effects model. High muscle Vitamin E improved redness in all samples across display, though to a lesser extent in 70 day aged lamb. High IMF worsened browning only in 5 day aged lamb, while high ICDH worsened browning after 35 days ageing, suggesting high IMF worsens browning in short aged lamb independent of oxidative capacity while Vitamin E supplementation is of greater benefit in short than long-aged lamb.

Key Words – Browning, color, muscle oxidative capacity.

I. INTRODUCTION

The rapid browning of lamb meat on retail display is an important problem for the Australian lamb industry as it deters consumers who associate brown meat with a lack of freshness and reduced quality [1]. Anecdotal evidence suggests that overwrapped lamb meat has a retail shelf life of only 2 days due to browning, when retailers are forced to discount otherwise high quality lamb to ensure it is sold. This brown colour change is caused by the accumulation of free radical species that oxidise myoglobin pigments into brown metmyoglobin [1].

A key factor influencing the rate of lamb browning is ageing of meat prior to retail display. Previous research has focused on shorter ageing periods, with browning rates becoming progressively more rapid for ageing periods up to 35 days in lamb [2,3,4]. However in practice lamb meat if often aged for up to 70 days when exported to distant markets such as the USA. Thus it is essential to determine whether this extended ageing further increases the rate of browning when cut for retail display.

This ageing-linked increase in the rate of lamb browning is likely related to increasing lipid peroxidation [5] and declining mitochondrial activity [6]. High lipid peroxidation increases the accumulation of oxidative free radicals [5] while reduced mitochondrial activity will result in reduced free radicals byproducts of oxidative metabolism. Other related intrinsic muscle factors may also impact the oxidative load in long-aged meat, including high levels of intramuscular fat, increased muscle aerobic capacity (indicated by isocitrate dehydrogenase activity; ICDH) and high ultimate pH [7,8]. A high pH favours increased mitochondrial activity, while high IMF is associated with both an increased lipid peroxidation as well as with high ICDH and consequently increased mitochondrial density [6]. These impacts have only been demonstrated in short-aged meat (up to 35 days), yet their effects may be even more pronounced in long-aged product (up to 70 days) where the inherent reducing capacity of the meat is compromised.

One plausible means of reducing the accelerated browning caused by long ageing, high IMF, ICDH and pH is to supplement lamb diets with the antioxidant Vitamin E prior to slaughter. Yet the

impact these intrinsic muscle factors on meat colour has only been shown in short-aged lamb meat [2,8]. High intramuscular Vitamin E is thought to improve meat colour by neutralising oxidative free radicals that instigate myoglobin oxidation [4]. Thus the capacity of Vitamin E to delay browning in lamb may be of greater magnitude in long-aged meat with high IMF, ICDH and pH given the greater free radical load anticipated in this meat.

Therefore we hypothesise that high intramuscular Vitamin E concentration in lamb *longissimus* will reduce browning (measured as redness) during display more in long-aged than short-aged lamb meat. In addition the effect of Vitamin E will be greater in lamb meat with a high intramuscular fat, high ICDH and with a high ultimate pH.

II. MATERIALS AND METHODS

Lambs (n=132) were selected from the Sheep Cooperative Research Centre's Resource Flock in Katanning, Western Australia. The lambs were the progeny of Merino and Border-Leicester Merino dams artificially inseminated by 66 industry sires with established breeding value estimates. The sires were a mix of Terminal, Maternal and Merinos and were selected based on sire breeding values for fat (PFAT; post-weaning fat depth) and muscling (PEMD; post-weaning eye muscle depth) to obtain variation among the progeny lambs in IMF (associated with PFAT) and muscle oxidative capacity as indicated by ICDH (associated with PEMD).

Two progeny from each sire were selected and randomly divided into two treatment groups (n=66); Low (30mg/kg feed) and High (275mg/kg feed) Vitamin E supplementation. The lambs were acclimatized to pellets whilst on pasture before being divided into 12 pens (6 pens per treatment), each containing 11 lambs with similar body weights representing a range of sires. The lambs were fed treatment rations for 8 weeks prior to slaughter at a mean carcass weight of 23 kg (± 0.3 SE).

At 24 hours post-mortem the m. *longissimus* was sampled for measurement of Vitamin E concentration, IMF, ICDH and pH (pH_{24}). 3 large samples of loin were collected per lamb to be vacuum packaged and aged at -1°C for 5, 35 and 70 days. Following ageing the samples were sliced to create a fresh meat surface before being wrapped with oxygen-permeable film and placed under simulated retail display for a period of 3 days. Surface redness was measured at intervals of 24 hours during display using a Hunterlab reflectometer. Redness was calculated using light reflectance at 630/580nm and was analysed using a mixed linear effects model in SAS, with ageing period, sire type, dam breed and gender as fixed effects, display time, Vitamin E, IMF, ICDH and pH_{24} as covariates, and sire and dam as random terms. Non-significant interactions (P>0.05) were removed in a step-wise fashion.

III. RESULTS AND DISCUSSION

Vitamin E concentrations ranged from 0.64 to 4.54 mg/g of muscle across the Vitamin E treatment groups. High intramuscular Vitamin E was associated with increased redness in 5, 35 and 70 day aged lamb loin during display (P<0.05). However contrary to our hypothesis the improvement in redness with high Vitamin E was slightly less in long-aged than short or medium aged meat. Increasing Vitamin E from 1 to 3.5 mg/g of muscle produced a 0.52, 0.68 and 0.44 unit increase in redness after 72 hours display for 5, 35 and 70 day aged loin meat respectively. The apparent reduced efficacy of Vitamin E supplementation in 70 day aged lamb suggests that the anti-oxidant capacity of Vitamin E is becoming overwhelmed by the substantial oxidative load in such long-aged meat.

Increasing ICDH across a range of 2 to 5 µmol/min/ g of muscle reduced redness by 1 unit after display (P<0.05) in 35 day aged meat. This association is likely related to increased mitochondrial quantity and thus activity, which is then irrelevant in very long-aged meat due to a loss of mitochondrial structural integrity and thus activity [6]. The lack of association between ICDH and redness in short-aged lamb contrasts with previous work [8] and is difficult to explain.

High IMF did increase the rate of meat browning, however only in short-aged lamb meat (P<0.05). In loin aged for 5 days increasing IMF from 2.5 to

5.5 % reduced redness at the end of display by 0.4 units, while changes in IMF did not alter redness during display after 35 or 70 days ageing. These results suggest that the negative impact of IMF on lamb browning may be confined to short-aged meat. The lack of association between IMF and browning in long-aged lamb suggests that any IMF-linked increase in lipid peroxidation and thus free radical production is negligible when compared to the overwhelming increase in lipid peroxidation and oxidative load in long-aged lamb.

The fact that IMF hastened the rate of browning in short-aged lamb while ICDH did not suggests that the impact of IMF in this meat is unrelated to any associated increase in muscle oxidative capacity.

The only effect of pH_{24} on browning was seen in 35 day aged meat where increasing pH_{24} from 5.5 to 5.8 increased redness by 1.0 unit after 72 hours on display. This unexpected result is contrary to the majority of research which describes a negative impact of high pH on browning in short-aged lamb [5,6]. This result may relate to an increase in reducing capacity that has been reported in high pH meat [9].

Contrary to our hypothesis the improvement in redness caused by high intramuscular Vitamin E was no greater in high IMF, ICDH or pH_{24} lamb meat. This result is unsurprising however given the lack of association between redness and IMF, ICDH and pH_{24} in this study, particularly in long-aged meat.

At a low IMF of 2.5% (Fig. 1) increasing Vitamin E from 1 to 3.5mg/g of muscle increased redness ($P<0.05$) after 72h display in 5, 35 and 70 day aged meat by 0.79, 0.90 and 0.33 units. Increasing IMF to 5.5% (Fig. 2) depressed this effect ($P<0.05$), with high Vitamin E only increasing redness by 0.69, 0.59 and 0.16 units (Fig. 2). In either case Vitamin E was less effective in 70 d aged meat ($P<0.05$).

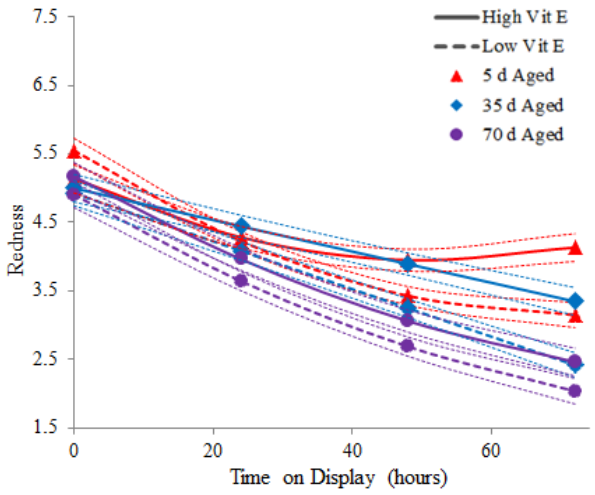

Figure 1. Effect of display time (hrs) on 2.5% IMF lamb m. *longissimus* redness. Lines represent least square means for each ageing period at high (3.5mg/g muscle) and low (1mg/g muscle) Vitamin E.

An interesting additional finding was the increase in meat redness ($P<0.05$) seen at the start of display (0 – 24 hours) in short-aged lamb with high Vitamin E (Fig. 2). The reason for this is unclear but may relate to Vitamin E increasing the bloom depth and thus initial red colour development of the meat. This is a positive result suggesting that Vitamin E supplementation is worthwhile to improve the colour of short-aged lamb throughout its retail display.

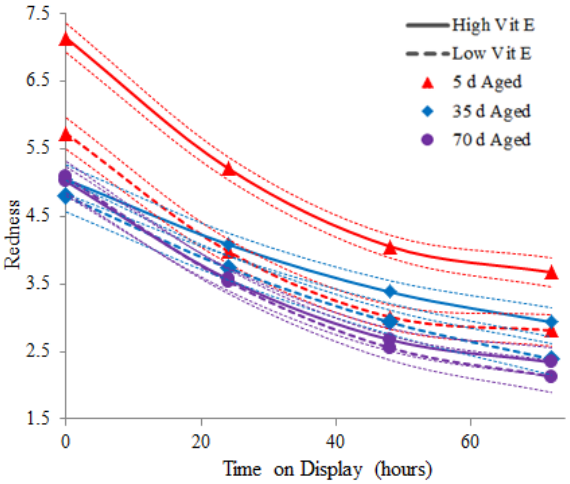

Figure 2. Effect of display time (hrs) on 5.5% IMF lamb m. *longissimus* redness. Lines represent least square means for each ageing period at high (3.5mg/g muscle) and low (1mg/g muscle) Vitamin E.

IV. CONCLUSION

The reduction in browning on display in medium and long-aged lamb meat with high Vitamin E concentrations in this study suggests that Vitamin E is a worthwhile supplement for Australian lamb destined for export. However Vitamin E supplementation may be of most value in short-aged domestic lamb product, where high muscle Vitamin E demonstrated the greatest impact improving redness throughout the retail display of highly marbled lamb loin.

ACKNOWLEDGEMENTS

The authors greatly acknowledge the financial support of the Cooperative Research Council for Sheep Industry Innovation and the large number of people who assisted at the Great Southern Agricultural Research Institute, at WAMMCO abattoir and at Murdoch University.

REFERENCES

1. Faustman, C., & Cassens, R.G. (1990). The biochemical basis for discoloration in fresh meat: A review. Journal of Muscle Foods 1(3): 217-243.
2. Jose, C., Gardner, G., Pethick, D. & Jacob, R. (2008). The colour stability of Aged lamb benefits from Vitamin E supplementation. In proceedings of 54th International Congress of Meat Science & Technology (p149), 10-15 August 2008, Cape Town, South Africa.
3. King, D.A., Shackelford, S.D. Kalchayanand, N. & Wheeler, T.L. (2012). Sampling and ageing effects on beef longissimus color stability measurements. Journal of Animal Science. 90: 3596-3605.
4. Wulf, D.M., Morgan, J.B., Sanders, S.K., Tatum, J.D., Smith, G.C. & Williams, S. (1995). Effects of dietary supplementation of Vitamin E on storage and caselife properties of lamb retail cuts. Journal of Animal Science. 73: 399-405.
5. Faustman, C., Sun, Q., Mancini, R. & Suman, S.P. (2010). Myoglobin and lipid oxidation interactions: Mechanistic bases and control. Meat Science 86: 86-94.
6. Tang, J., Faustman, C., Hoagland, T.A., Mancini, R.A., Seyfert, M. & Hunt, M.C. (2005). Postmortem oxygen consumption by mitochondria and its effects on myoglobin forma and stability. Journal of Agriculture and Food Chemistry 53: 1223-1230.
7. Jacob, R.H., D'Antuono, M.F., Gilmour, A.R. & Warner, R.D. (2014). Phenotypic characterization of colour stability of lamb meat. Meat Science 96: 1040-1048
8. Calnan, H.B., Jacob, R.H., Pethick, D.W., & Gardner, G.E. (2014). Factors affecting the colour of lamb meat from the *longissimus* muscle during display: The influence of muscle weight and muscle oxidative capacity. Meat Science 96: 1049-1057.
9. Ledward, D.A. (1985) Post-slaughter influences on the formation of metmyoglobin in beef muscles. Meat Science 15:149-171.