# Effect of extended storage on the quality and stability of conjugated linoleic acid (CLA) in wet and dry-aged, frozen-thawed beef

Tanyaradzwa E Mungure[1*], Alaa El-Din Bekhit[1], John E Birch[1], Dong Ju Kim[1], Alan Carne[2], Ian Stewart[3] and Mustafa M Farouk[4]

[1]Department of Food Science, University of Otago, Dunedin, New Zealand; [2]Department of Biochemistry, University of Otago, Dunedin, New Zealand; [3]Department of Chemistry, University of Otago, Dunedin, New Zealand; [4]AgResearch, Ruakura Research Centre, P Bag 3115, Hamilton, New Zealand;

*Corresponding author email: tanyaradzwa.mungure@postgrad.otago.ac.nz

**Abstract – This study determined the effect of extended post-ageing storage of dry vs. wet aged, frozen-thawed beef. Strip-loins were dry or wet aged for 21 days, vacuum-packed, and stored at -20°C for 24 months. Samples were analysed for tenderness, lipid oxidation and the stability of bioactive CLA. $^1$HNMR spectroscopy was applied to assess lipid oxidation. Dry aged samples had higher $R_{ad}$ and $R_{ao}$ ratios ($p < 0.05$), decreased PUFA, MUFA and PUFA/SFA ratios ($p < 0.05$) indicating higher lipid oxidation. Dry aged samples had higher CLA concentrations ($p < 0.05$), but no difference in tenderness compared to wet aged ($p > 0.05$) were found. Comparing the two ageing regimes and extended storage, dry ageing did not negatively affect CLA or tenderness.**

**Key Words – lipid oxidation, $^1$H NMR, tenderness.**

## I. INTRODUCTION

Dry ageing of beef involves ageing of premium cuts under controlled ambient conditions, temperature and airflow [1]. Compared to wet (vacuum packed) aged meat, it has a characteristically unique flavour. Several studies have been done to understand the effects of dry ageing on meat quality attributes, but with limited work done on the implications of long storage post-ageing on meat quality, lipid oxidation and the stability of CLA. CLAs consist of a group of geometric and positional isomers of linoleic acid and have been reported to provide antidiabetic, anti-atherogenic and anticancerogenic effects [2]. This study analyses the effects of long-term storage on meat quality attributes and CLA stability for wet and dry-aged beef. Long storage of dry aged premium cuts may facilitate extended supply and better supply chain control.

## II. MATERIALS AND METHODS

*Raw materials and processing*

A total of 10 loin sections (*M. longissimus lumborum*; bone-in) were split into two treatments: wet (under vacuum packaging) and dry aged. They were stored at 3°C with an air velocity of 0.2m $s^{-1}$ for 21 days [3]. The samples were then stored at -20°C for 24 months. Cooking loss and shear force was determined as reported by Mungure et al. [4]. Lipid extraction for oxidation and CLAs analyses was according to Folch [5]. Fatty acid methyl esterification (FAME) production from lipid and analysis by GC-FID was performed as reported by Mungure et al. [4]. $^1$H NMR was used to analyse lipid oxidation via the depletion of polyunsaturated fatty acids (PUFA) and monounsaturated fatty acid acids (MUFA) using the $R_{ad}$ (aliphatic to diallylmethylene/ bisallylic proton) ratio and $R_{ao}$ (aliphatic to olefinic proton) ratio. $^1$ H NMR was also used to quantitate CLAs using modified method of Prema et al. [2] respectively.

## III. RESULTS AND DISCUSSION

*pH, thawing, cooking loss, shear force*

The ageing regimes did not have a significant influence on the pH ($p > 0.05$), however as shown in table 1, the overall mean of both treatments were quite high. This could be attributed to meat protein degradation by endogenous enzymes leading to the production of organic alkaline sulphides, amines and ammonia increasing the pH. Thawing and cooking loss were both significantly higher in wet compared to the dry aged samples ($p < 0.05$). There was no significant effect on shear force ($p > 0.05$).

Table 1 The mean values for meat quality, lipid oxidation and CLA from the wet and dry aged beef after 2 years of storage time.

| Ageing treatment | pH | Thawing loss (%) | Cooking loss (%) | Shear force (N) | $R_{ad}$ | $R_{ao}$ | CLA (mg/g lipid) | CLA (%FA) |
|---|---|---|---|---|---|---|---|---|
| Wet | 5.95$^a$ | 1.82$^a$ | 30.80$^a$ | 42.21$^a$ | 22.84$^b$ | 54.18$^b$ | 1.85$^b$ | 1.08$^a$ |
| Dry | 5.83$^a$ | 0.76$^b$ | 26.45$^b$ | 36.73$^a$ | 29.77$^a$ | 75.39$^a$ | 2.70$^a$ | 0.66$^b$ |
| SEM | 0.07 | 0.17 | 1.31 | 3.29 | 0.86 | 3.27 | 0.16 | 0.06 |

$^{ab}$Figures with different letter in a column are significant different (at $p < 0.05$)

*Lipid oxidation ($R_{ao}$, $R_{ad}$ ratios and fatty acid profiles) and CLA analysis*

$R_{ad}$ ratio was significantly higher in dry aged compared to the wet ($p < 0.05$), revealing higher oxidation. This observation was supported by the fatty acid profiles (table 2). PUFA levels in dry aged beef were significantly lower than wet ($p < 0.05$), and the PUFA/SFA ratio was higher for wet aged compared to dry (table 2).

Table 2 the mean values for fatty acid profiles in wet and dry aged beef after 2 years of storage time.

| Fatty acid | C14:0 | 14:1 | C16:0 | C16:1 | C17:0 | C17:1 | C18:0 | C18:1tω-9 |
|---|---|---|---|---|---|---|---|---|
| Wet aged (% FA) | 2.80 ± 0.33[a] | 0.66±0.30[a] | 26.18±0.34[b] | 4.57±0.37[a] | 0.54±0.14[b] | 0.49±0.18[a] | 13.56±0.84[b] | 2.18±0.17[a] |
| Dry aged (% FA) | 2.35 ± 0.33[a] | 0.70±0.30[a] | 27.63±0.34[a] | 3.17±0.37[b] | 1.06±0.14[a] | 0.54±0.18[a] | 18.80±0.84[a] | 0.95±0.17[b] |
| Fatty acid | C18:1c ω-9 | C18:1t ω-7 | C18:2t ω-6 | C18:2c ω-6 | C18:2 CLA | C18:3 ω-3 | C18:3 ω-6 | C20:0 |
| Wet aged (% FA) | 42.46±0.48[a] | 1.37±0.09[a] | 0.37±0.11[a] | 2.27±0.09[a] | 0.66±0.06[b] | 1.15±0.03[a] | 0.28±0.04[a] | 0.35±0.05[a] |
| Dry aged (% FA) | 39.35±0.48[b] | 1.14±0.09[a] | 0.29±0.11[a] | 1.55±0.09[b] | 1.08±0.06[a] | 0.78±0.03[b] | 0.07±0.04[b] | 0.48±0.05[a] |
| Fatty acid | C20:1 | SFA | MUFA | PUFA | TUFA | PUFA/SFA | | |
| Wet aged (% FA) | 0.12±0.04[a] | 43.43±0.57[b] | 51.85±0.60[a] | 4.72±0.15[a] | 56.57±0.57[a] | 0.10[a] | | |
| Dry aged (% FA) | 0.06±0.04[a] | 50.32±0.57[a] | 45.92±0.60[b] | 3.77±0.15[b] | 49.68±0.57[b] | 0.07[b] | | |

[ab]Fatty acids % with different letter in a column are significantly different (at $p < 0.05$)

Dry aged meat had higher $R_{ao}$ ratio compared to wet ($p < 0.05$), highlighting the decline in the monounsaturated lipids showing lipid oxidation. This is supported by the fatty acid profile above. The average C18:1*c* ω-9 (a MUFA) was significantly lower in dry vs. wet aged meat ($p < 0.05$).

Dry aged meat had a significantly higher CLA concentration compared to wet ($p < 0.05$). The concentrations across all samples were in the range of 1.2-3.2mg/g of beef lipid. All dry aged samples had concentrations in excess of 2.3mg/g lipid. CLA has been reported to be more stable than other PUFAs that contain methylene interrupted double bonds, this leads to PUFAs being more susceptible to oxidation. This data shows depletion of CLA does not necessarily follow the same pathway as PUFAs.

## IV. CONCLUSION

The implication of this study is that dry aged beef can be stored frozen for 24 months with no deleterious effect on CLA concentrations and tenderness compared to wet aged beef. Lipid oxidation is however higher in dry aged beef after the storage time.

## ACKNOWLEDGEMENTS

T Mungure thanks AgResearch Hamilton NZ for funding this project and the University of Otago for a doctoral scholarship.

## REFERENCES

1. DeGeer, S., Hunt, M., Bratcher, C., Crozier-Dodson, B., Johnson, D., & Stika, J. (2009). Effects of dry aging of bone-in and boneless strip loins using two aging processes for two aging times. *Meat Science, 83*(4), 768-774.
2. Prema, D., Turner, T. D., Jensen, J., Pilfold, J. L., Church, J. S., Donkor, K. K., & Cinel, B. (2015). Rapid determination of total conjugated linoleic acid concentrations in beef by 1H NMR spectroscopy. *Journal of Food Composition and Analysis, 41*, 54-57.
3. Kim, Y. H. B., Kemp, R., & Samuelsson, L. M. (2016). Effects of dry-aging on meat quality attributes and metabolite profiles of beef loins. *Meat Science, 111*, 168-176.
4. Mungure, T. E., Bekhit, A. E.-D. A., Birch, E. J., & Stewart, I. (2016). Effect of rigor temperature, ageing and display time on the meat quality and lipid oxidative stability of hot boned beef Semimembranosus muscle. *Meat Science, 114*, 146-153.
5. Folch, J., Lees, M., & Sloane-Stanley, G. (1957). A simple method for the isolation and purification of total lipids from animal tissues. *J Biol Chem, 226*(1), 497-509.