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The effects of graded levels of concentrate supplementation on colour and lipid stability of beef from pasture finished late-maturing bulls

S. Siphambili[1,2], A. P. Moloney[2], E. G. O'Riordan[2], M. McGee[2] and F. J. Monahan[1†]

[1]*UCD School of Agriculture and Food Science, University College Dublin, Belfield, Dublin 4, Ireland;* [2]*Teagasc, Animal & Grassland Research and Innovation Centre, Grange, Dunsany, Co. Meath, Ireland*

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*Finishing late-maturing bulls on grass may alter the antioxidant/prooxidant balance leading to beef with higher susceptibility to lipid oxidation and a lower colour stability compared to bulls finished on cereal concentrates. In this context, lipid oxidation and colour stability of beef from late-maturing bulls finished on pasture, with or without concentrate supplements, or indoors on concentrate was assessed. Charolais or Limousin sired bulls (*n = 48*) were assigned to four production systems: (1) pasture only (**P**), (2) pasture plus 25% dietary DM intake as barley-based concentrate (**PC25**), (3) pasture plus 50% dietary DM intake as barley-based concentrate (**PC50**) or (4) a barley-based concentrate ration (**C**). Following slaughter and* postmortem *ageing,* M. Longissimus thoracis et lumborum *was subjected to simulated retail display (4°C, 1000 lux for 12 h out of 24 h) for 3, 7, 10 and 14 days in modified atmosphere packs (O_2 : CO_2; 80 : 20). Lipid oxidation was determined using the 2-thiobarbituric acid-reactive substances assay; α-tocopherol was determined by HPLC; fatty acid methyl esters were determined using Gas Chromatography. Using a randomised complete block design, treatment means were compared by either ANOVA or repeated measures ANOVA using the MIXED procedure of SAS. Total polyunsaturated fatty acid (**PUFA**) concentrations were not affected by treatment,* n-3 *PUFAs were higher (*$P < 0.001$*) and the ratio of* n-6 *to* n-3 *PUFAs was lower (*$P < 0.001$*) in muscle from P, PC25 and PC50 compared to C. α-Tocopherol concentration was higher in muscle from P compared to PC50 and C bulls (*$P = 0.001$*) and decreased (*$P < 0.001$*) in all samples by day 14. Lipid oxidation was higher in muscle from C compared to P bulls on day 10 and day 14 of storage (*$P < 0.01$*). Finishing on pasture without supplementation did not affect beef colour stability and led to lower lipid oxidation, possibly due to the higher α-tocopherol concentration compared to concentrate finished beef.*

Keywords lipid oxidation, fatty acids, polyunsaturated fatty acids, α-tocopherol, shelf life

Implications

The diet during the finishing period of bull beef production systems can affect muscle retail shelf life as a result of changes in the fatty acid and antioxidant concentration. Pasture finishing of late-maturing bulls results in a lean product with high antioxidant (α-tocopherol) concentration such that beef from bulls finished at pasture is more stable than concentrate-fed beef despite a higher proportion of polyunsaturated fatty acids.

Introduction

The production of beef from intact males is mainly based on feeding cereal-based concentrates *ad libitum* or conserved forage supplemented with concentrates (O'Riordan *et al.*, 2011), with slaughter at between 12 and 16 months, but does not usually include grazed grass as part of the growing or finishing phase. The cost of concentrates can be difficult to offset when beef prices are low or when feed ingredient prices are high. Grazed grass, which is relatively low in cost compared to concentrates (Finneran *et al.*, 2012), is available in abundance in countries with temperate climates. It has been shown to reduce the cost of suckler bull beef production (O'Riordan *et al.*, 2011). Lower energy intake on grass-based diets may have an impact on fat deposition, with low fat deposition affecting consumer acceptability. Intramuscular fat (**IMF**) has been shown to be positively correlated with beef tenderness, juiciness, flavour and overall palatability (Mezgebo *et al.*, 2017a).

† E-mail: frank.monahan@ucd.ie

The finishing of bulls on grass may result in a leaner product with a higher ratio of polyunsaturated fatty acids (**PUFAs**) to saturated fatty acids (**SFAs**) compared to concentrate finishing. This may have repercussions for retail shelf life due to changes in the prooxidant/antioxidant balance. The resistance of muscle to lipid oxidation depends on the proportion of prooxidant (e.g. PUFAs) to antioxidant (e.g. α-tocopherol) components (Morrissey *et al*., 1994). Luciano *et al*. (2011) showed that grass-fed heifers, including those given 50% dietary DM of concentrate, had a 1.3-fold higher highly peroxidisable PUFA (**HP-PUFA**) concentration and 1.3- to 1.6-fold higher PUFA proportion in muscle compared to concentrate-fed heifers. Bulls tend to have lower IMF deposition compared to heifers or cows (Chriki *et al*., 2013) and steers (Purchas *et al*., 2002). Therefore, finishing late-maturing bulls on pasture may present a challenge to lipid stability due to the increased proportion of PUFAs in muscle.

It was hypothesised that there would be differences in the susceptibility to oxidation of beef from late-maturing bulls fed graded levels of concentrates at pasture due to differences in the prooxidant to antioxidant balance.

Materials and methods

Animals, diets and management

Forty eight autumn-born, Charolais or Limousin sired crossbred suckler bulls, approximately 12 months old, were assembled in October/November and offered high nutritive value grass silage *ad libitum* plus 1.7 kg/head daily of a concentrate ration during the wintering period (135 days) (Lenehan, 2016). The bulls were blocked according to weight, age and breed and, from within block, randomly assigned to one of four production systems (**PS**) with 12 animals per production system. Three of the PS were pasture-based in which bulls rotationally grazed on perennial ryegrass (*Lolium perenne*) dominant swards: pasture only (**P**), pasture plus 25% dietary intake on a DM basis of a rolled barley-based concentrate (**PC25**) or pasture plus 50% dietary DM intake of the barley-based concentrate (**PC50**). The fourth PS was the barley-based concentrate offered *ad libitum* to housed bulls (**C**). The bulls on pasture supplemented with concentrate were offered the concentrates in one feed each morning in plastic troughs (0.5 m feed space allowance per animal). The composition of the barley-based concentrate is given in Table 1. Grass silage was offered *ad libitum* to animals on the concentrate diet (C). The concentrate allowance was gradually increased over 21 days until available *ad libitum*. The average DM intake (**DMI**) at pasture was estimated using the herbage disappearance method without correction for concurrent grass growth in the estimation. Grass samples for fatty acid (**FA**) and α-tocopherol analysis were taken from material cut from 5 m long strips (four strips) in each paddock and dried for 48 h at 40°C. Representative samples of silage and concentrates were collected once weekly at feeding and stored at –18°C prior to FA and α-tocopherol analysis. Further information on the management of animals and feed is given in Lenehan (2016).

Table 1 *Composition of the concentrate ration (g/kg fresh weight) used in the bull experimental diets*

Rolled barley	862
Soya bean meal	60
Cane molasses	50
Minerals and vitamins[a]	28

[a]Vitamin A 2 000 000 IU/kg, Vitamin D_3 400 000 IU, Vitamin E (α-tocopherol acetate) 6000 mg/kg, Vitamin B_1 1000 mg/kg, Vitamin B_{12} 3 mg/kg, Iron 4000 mg/kg, iodine 1000 mg/kg, cobalt 200 mg, copper sulphate 2000 mg/kg, copper chelate 2000 mg/kg, manganese 10 000 mg/kg, zinc 20 000 mg/kg, selenium 100 mg/kg (David Taylor, Animal Nutrition Ltd, Carrick Mill, Lough Bawn, Collinstown, Co. Westmeath, Ireland).

Treatments were applied for 120 days after which all animals were slaughtered at approximately 19 months of age. The bulls were slaughtered at a commercial abattoir (Kepak Group, Clonee, Co. Meath, Ireland) in two slaughter events with treatments equally distributed across slaughter events.

Sample collection

The carcasses were placed in a chill at 8°C to 10°C for 10 h *post-mortem* and then at 0°C up to 48 h *post-mortem* before muscle samples were collected. Samples of the *M. Longissimus thoracis et lumborum* were excised from between the 6th and 10th rib and dissected free of subcutaneous and intermuscular adipose tissue. After the removal of a 2.53 cm thick slice for proximate analysis (stored at –20°C), the muscle was vacuum-packed and aged for 14 days in total. After ageing the muscle was cut into individual 2.53 cm thick samples. Two samples were cut to give five different subsamples (two from one steak, three from the other). The samples were individually packed in polyamide-polyethylene Exovac 73 bags (McDonnells, Dublin, Ireland), using a Webomatic vacuum-packaging system equipped with a gas mixer (Witt-Gase Technik KM100-M(3) Witt Gas Techniques Ltd, Warrington, UK) to give modified atmosphere packs (**MAP**, 80% O_2 : 20% CO_2). The five subsamples were randomly assigned to simulated retail display (4°C, 1000 lux for 12 h out of 24 h) for 0, 3, 7, 10 and 14 days. The gas composition of the MAP was checked at the end of each display period using a hand-held gas analyser (PBI Dansensor Checkpoint, Ringsted, Denmark). At the end of each display period the samples were subdivided into three subsamples, for measurement of lipid oxidation, α-tocopherol and FAs and stored at –20°C in vacuum packs until further analysis.

Proximate analysis

Proximate analysis (moisture, intramuscular fat, protein) was undertaken following Association of Official Analytical Chemists methods (Supplementary Material S1).

Fatty acid composition

Fatty acid methyl esters (**FAMEs**) were prepared using the rapid microwave assisted method described by

Brunton *et al.* (2015) (Supplementary Material S1). The atherogenic index (**AI**), which is the relationship between the main saturated (proatherogenic) and the unsaturated fatty acids (antiatherogenic), was calculated using the following formula: (C12:0 + 4 × C14:0 + C16:0)/(∑PUFA + ∑MUFA). The thrombogenic index (**TI**), which estimates the potential to form clots in blood vessels, was calculated using the following formula: (C14:0 + C16:0 + C18:0)/(0.5 × ∑MUFA + 0.5 × *n*-6 + 3 × *n*-3 + *n*-3/*n*-6) (Ulbricht and Southgate, 1991).

α-Tocopherol concentration

Vitamin E (α-tocopherol) in feed was measured following the method of Fratianni *et al.* (2002) with minor modifications (Supplementary Material S1). α-Tocopherol in muscle was extracted as described in Dunne *et al.* (2005a).

Meat colour measurements

Colour measurements were made following American Meat Science Association procedures (American Meat Science Association, 2012). A Minolta Chroma Meter (CR-400, Konika Minolta, Mason Technology, Dublin, Ireland) was set at illuminant D65, 2° standard observer and Hunter colour scale 'L', 'a', 'b', with 'L' indicating lightness, 'a' indicating redness and 'b' indicating yellowness. For day 0 measurements the steaks were allowed to bloom for 90 min in the MAP before measurement. Colour co-ordinates were obtained from three different non-overlapping points on each sample through the film at 0, 3, 7, 10 and 14 days of simulated retail display.

Lipid oxidation

Lipid oxidation was estimated by measuring 2-thiobarbituric acid-reactive substances (**TBARS**) as described by Luciano *et al.* (2011). A calibration curve was prepared using known concentrations of 1, 1, 3, 3-tetra-ethoxypropane in distilled water. The absorbance at 532 nm was measured using a Shimadzu UV Mini 1240 spectrophotometer (Mason Technology, Dublin, Ireland).

Statistical analysis

The experimental design was a randomised complete block design with animal as the experimental unit. Analysis of variance and repeated measures ANOVA using the MIXED procedure of SAS (Version 9.4, SAS Institute Inc., Cary, NC, USA) were used to compare the treatments. For proximate analysis, where there was one observation per animal, PS and block were treated as fixed effects while the animal was treated as a random effect. For repeated measures (FA composition, α-tocopherol concentration, colour and lipid oxidation) PS, storage time, block and their interactions were treated as fixed effects while the animal was treated as a random effect. Data were checked for normality using the Proc Univariate procedure of SAS, and non-normal data were transformed. Differences between least square means were determined using Bonferroni adjustment for multiple comparisons at a significance level of $P < 0.05$ and considered a tendency when $P > 0.05$ but < 0.10. Least square means are reported with pooled standard errors (SEM).

Results

Feed fatty acid composition and α-tocopherol concentration

The FA composition and α-tocopherol concentration of feeds are shown in Table 2. The average total FA concentration was higher in grass than in concentrate and silage (Table 2). The mean SFA concentration was higher in concentrate than in silage and grass. The mean MUFA concentration was about fourfold higher in concentrate than in silage and grass. Silage and grass had 1.5- and 1.7-fold higher PUFA, respectively, than concentrate. The mean *n*-6 PUFA concentration of concentrate was about threefold higher than that of grass silage and grass. The *n*-3 PUFA concentration of silage and grass was 19.2- and 23.4-fold higher, respectively, than that of

Table 2 *Fatty acid and α-tocopherol concentration in silage, grass and concentrate (DM basis) fed to late maturing bulls*

	Concentrate	Grass	Silage
	Mean (SD)	Mean (SD)	Mean (SD)
	n = 6	*n* = 16	*n* = 6
Fatty acids mg/kg DM			
C12	23.5 (19.9)	50.3 (11.9)	346 (159)
C14	88.4 (18.4)	129 (12.2)	164 (56.1)
C14:1	0.0	6.86 (6.36)	10.9 (7.97)
C15:0	11.2 (7.28)	34.3 (9.26)	44.0 (7.85)
C15:1	8.51 (1.62)	6.14 (1.45)	7.65 (1.49)
C16:0	4813 (450)	3539 (253)	3017 (492)
C16:1	21.8 (3.28)	84.7 (45.6)	65.9 (21.9)
C17:0	19.8 (1.27)	39.6 (5.59)	35.9 (4.64)
C18	400 (37.1)	462 (53.3)	326 (76.5)
C18:1*n*-9	2512 (223)	514 (52.9)	372 (37.9))
C18:1*n*-7	185 (19.1)	133 (38.2)	166 (42.2)
C18:2*n*-6*c*	6167 (552)	2026 (267)	2236 (319)
C20:0	38.0 (4.81)	96.7 (8.02)	122 (14.5)
C20:1	108 (10.3)	23.3 (5.27)	22.0 (6.21)
C18:3*n*-3	391 (42.8)	8985 (2032)	7267 (2006)
C:21	27.8 (2.93)	–	–
C20:2	13.1 (1.28)	3.40 (4.66)	7.25 (3.96)
C22:0	46.7 (10.3)	218 (14.5)	230 (86.4)
C20:3*n*-3	0.0	145 (19.4)	179 (33.6)
C22:2	0.0	18.7 (5.19)	16.0 (6.44)
C24:0	40.8 (3.05)	191 (11.8)	216 (76.6)
Others	280 (19.6)	280 (22.5)	401 (93.3)
Total fatty acids	15 196 (1231)	17 067 (2592)	15 250 (3266)
SFA	5510 (483)	4848 (319)	4499 (924)
MUFA	2835 (123)	761 (128)	645 (40.9)
PUFA	6571 (594)	11 178 (2300)	9704 (2345)
n-6 PUFA	6180 (552)	2026 (267)	2259 (323)
n-3 PUFA	391 (42.7)	9130 (2030)	7445 (2032)
α-Tocopherol mg/kg DM	15.8 (1.57)	21.1 (6.58)	64.9 (22.5)

SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acid.

Table 3 *Proximate composition (g/kg) of M. longissimus thoracis et lumborum muscle of late-maturing bulls from four production systems*

	Production system					
	C	P	PC25	PC50	SEM	*P*-value
IMF	14.6[b]	1.6[a]	2.4[a]	2.6[a]	1.42	***
Protein	230	228	230	226	1.67	n.s.
Moisture	741[a]	757[b]	753[b]	754[b]	1.73	***
Ash	10.7	10.8	11.1	10.9	0.19	n.s.

C = concentrate; P = grass only; PC25 = grass with 25% concentrate DM; PC50 = grass with 50% concentrate DM; IMF = intramuscular fat content.
[a,b]Least square means within rows assigned different superscripts differ significantly, $P < 0.05$.
*** $P < 0.001$.

concentrate. The mean α-tocopherol concentration of silage was threefold higher than that of grass and four-fold higher than that of concentrate (Table 2).

Proximate composition of muscle

The proximate composition of muscle from the bulls in the different PS is shown in Table 3. The IMF content of muscle from C was higher ($P < 0.001$) than muscle from P, PC25 and PC50 which did not differ. The protein content of muscle was not significantly different ($P > 0.05$) between PS. The moisture content was lower ($P < 0.001$) in muscle of C compared to P, PC25 and PC50 bulls which did not differ. Ash content was not significantly different between PS.

Muscle fatty acid concentration

There was no interaction between PS and storage time for any FA; therefore, only the effects of PS and storage time on muscle FA expressed as mg/100 g of muscle are shown in Table 4. The concentrations of C10:0 and C12:0 in muscle from C bulls were higher ($P < 0.001$) compared to P and PC50 bulls while muscle from PC25 bulls had similar concentrations ($P > 0.05$) to C, P and PC50. The concentrations of C14:0, C14:1, C15:0, C16:0, C16:1, C17:0, C18:0, C18:1*n*-9*c*, C18:1*n*-7, C20:1*n*-9 SFA, C20:2, total FA, SFA and MUFA in muscle were higher ($P < 0.001$) in C compared to P, PC25 and PC50 bulls, which did not differ. The concentrations of C15:1, C17:1, C18:2*n*-6*t*, C20:0, C18:3*n*-6, C18:2*c*9*t*11, C22:0, C20:3*n*-6, C20:3*n*-3, C20:4*n*-3, C22:2, C24:0 and PUFA were not affected ($P > 0.05$) by PS. The concentrations of C18:2*n*-6*c* and total *n*-6 PUFA in muscle from P and PC25 bulls were lower ($P < 0.001$) compared to C bulls, while concentrations in muscle from P, PC25 and PC50 bulls were similar ($P > 0.05$). The concentrations of C18:2*n*-6*c* and total *n*-6 PUFA in muscle from PC50 and C bulls did not differ ($P > 0.05$). The concentrations of C18:3*n*-3, C20:5*n*-3, C22:6*n*-3, *n*-3 PUFA and HP-PUFA were lower ($P < 0.01$) in muscle from C than P, PC25 and PC50 bulls, which did not differ ($P > 0.05$). The PUFA : SFA ratio was lower ($P < 0.001$) in muscle from C compared to P, PC25 and PC50 bulls, which did not differ. The *n*-6 : *n*-3 ratio was higher ($P < 0.001$) in the muscle of C compared to P, PC25 and PC50 bulls and in PC50 compared to P but PC25 was similar ($P > 0.05$) to both P and PC50. The AI and TI were both higher ($P < 0.05$) in the muscle of C bulls compared to P, PC25 and PC50 bulls and increased ($P < 0.05$) with storage time.

When the FA data were expressed on a proportion basis (Supplementary Material S1), there was no interaction between PS and storage time. The effect of PS on FA was no longer significant for C12:0, C15:0, C18:1*n*-7, C20:1*n*-9 and C20:2, while it became significant for C15:1, C22:0, C20:3*n*-6, C22:2 and PUFA. The proportions of C14:0, C16:0, C16:1, C18:1*n*-9*c* and total MUFA were lower ($P < 0.001$) in muscle from P, PC25 and PC50 bulls, which did not differ, compared to C. The proportion of C14:1 was lower ($P < 0.001$) in P, PC25 and PC50 compared to C bulls and lower in muscle from P compared to PC50 bulls, whereas the proportion in muscle from PC25 did not differ ($P > 0.05$) from either P or PC50 bulls. The proportion of C15:1 was lower ($P < 0.001$) in C compared to P, PC25 and PC50 bulls, lower in P compared to PC25 bulls and did not differ between PC25 and PC50 or P and PC50 bulls. The proportions of C18:0, C18:2*c*9 *t*11, C18:2*n*-6*c*, C18:3*n*-3, C20:3*n*-6, C20:4*n*-6, C22:2, C20:5*n*-3, C22:5*n*-3, C22:6*n*-3, PUFA, HP-PUFA, *n*-6 PUFA and *n*-3 PUFA were higher ($P < 0.001$) in muscle from P, PC25 and PC50 compared to C bulls. The proportions of C18:3*n*-6 and C18:2*c*9 *t*11 were higher ($P < 0.007$) in muscle from P and PC50 compared to C bulls and did not differ between C and PC25 bulls. The proportions of C22:0 and C20:4*n*-3 were higher in muscle from P compared to C bulls ($P < 0.001$). The proportions of C18:3*n*-3, C22:0, C22:5*n*-3, *n*-3 PUFA and HP-PUFA were higher ($P < 0.001$) in muscle of P compared to PC25 and PC50 bulls, which did not differ.

Storage time had a significant effect on some FA concentrations (Table 4). The concentrations of C15:1, C18:2*n*-6*c*, C18:3*n*-6, C18:3*n*-3, C20:3*n*-6, C20:4*n*-6, C22:2, C20:5*n*-3, C22:5*n*-3, C22:6*n*-3, PUFA, *n*-6 PUFA, *n*-3 PUFA and HP-PUFA and the PUFA : SFA ratio were lower ($P < 0.01$) on day 14 compared to day 0. The *n*-6 : *n*-3 ratio was higher ($P = 0.001$) on day 14 compared to day 0. The concentration of C20:4*n*-6 and HP-PUFA decreased significantly across all PS during storage, while C20:3*n*-6, C20:5*n*-3 and PUFA significantly decreased only in C, PC25 and PC50 and significant decreases were only observed in PC25 and PC50 for C18:3*n*-3, C20:5*n*-3, C22:6*n*-3, *n*-6 PUFA and *n*-3 PUFA. The concentration of C18:2*n*-6*c* and C18:3*n*-6 only showed a significant decrease in the muscle from PC25 and PC bulls, respectively.

The proportions of SFA, MUFA, C15:0, C16:0, C18:0 and C18:1*n*-9*c* increased with an increase in day of storage (Supplementary Material S1). There was an overall decrease ($P < 0.05$) in the proportions of C15:1, C18:2*n*-6*c*, C18:3*n*-6, C18:3*n*-3, C20:3*n*-6, C20:4*n*-6, C22:2, C20:5*n*-3, C22:5*n*-3, C22:6*n*-3, PUFA, *n*-6 PUFA, *n*-3 PUFA and HP-PUFA with increase in storage time. With regard to PS, a significant decrease in muscle FA proportion was observed in PC25 where C22:5*n*-3 and HP-PUFA were significantly lower on day 14.

Table 4 *Fatty acid concentration of intramuscular lipid from* M. longissimus thoracis et lumborum *muscle of late-maturing bulls from four production systems (PS)*

	Day 0				Day 14[1]					*P*-values	
	C	P	PC25	PC50	C	P	PC25	PC50	SEM	PS	Day
Fatty acids composition mg/100 g											
C10:0	0.55^{b}	0.11^{a}	0.14^{ab}	0.05^{a}	0.79^{b}	0.04^{a}	0.17^{ab}	0.02^{a}	0.07	***	n.s.
C12:0	1.13^{b}	0.41^{a}	0.46^{ab}	0.28^{a}	1.36^{b}	0.27^{a}	0.46^{ab}	0.20^{a}	0.11	***	n.s.
C14:0	58.4^{b}	9.97^{a}	17.1^{a}	18.0^{a}	66.4^{b}	11.1^{a}	16.0^{a}	16.5^{a}	0.16	***	n.s.
C14:1	10.9^{b}	0.83^{a}	2.20^{a}	2.39^{a}	11.9^{b}	1.10^{a}	1.66^{a}	1.84^{a}	0.90	***	n.s.
C15:0	9.58^{b}	3.29^{a}	4.11^{a}	4.76^{a}	9.95^{b}	3.94^{a}	4.39^{a}	4.84^{a}	0.77	***	n.s.
C15:1	2.68	3.01	2.97	2.90	1.62	2.30	1.92	2.15	0.17	n.s.	***
C16:0	587^{b}	139^{a}	211^{a}	210^{a}	607^{b}	148^{a}	194^{a}	196^{a}	36.6	***	n.s.
C16:1	70.2^{b}	12.1^{a}	21.7^{a}	20.0^{a}	77.1^{b}	13.1^{a}	19.5^{a}	17.9^{a}	4.34	***	n.s.
C17:0	26.3^{b}	6.77^{a}	8.88^{a}	10.3^{a}	26.4^{b}	7.73^{a}	9.46^{a}	9.92^{a}	2.24	***	n.s.
C17:1	1.45	1.28	0.84	0.94	5.77	0.04	1.43	0.98	0.97	n.s.	n.s.
C18:0	351^{b}	131^{a}	158^{a}	169^{a}	386^{b}	140^{a}	152^{a}	160^{a}	23.7	***	n.s.
C18:1*n*-9*c*	819^{b}	170^{a}	254^{a}	248^{a}	887^{b}	190^{a}	232^{a}	225^{a}	54.2	***	n.s.
C18:1*n*-7	38.9^{b}	12.5^{a}	17.0^{a}	15.6^{a}	39.3^{b}	12.3^{a}	14.2^{a}	13.91^{a}	2.14	***	n.s.
C18:2*n*-6t	0.17	0.20	0.09	0.08	0.46	0.16	0.43	0.18	0.08	n.s.	*
C18:2*n*-6*c*	101^{b}	64.1^{a}	78.5^{aby}	85.1^{ab}	78.6^{b}	50.9^{a}	50.6^{ax}	59.8^{ab}	4.19	***	***
C20:0	2.47	1.23	1.32	1.73	2.57	0.98	2.57	1.87	0.49	n.s.	n.s.
C18:3*n*-6	0.44	0.31	0.45	0.61^{y}	0.28	0.34	0.20	0.27^{x}	0.05	n.s.	***
C20:1*n*-9	3.53^{b}	0.86^{a}	1.47^{a}	1.18^{a}	3.40^{b}	0.90^{a}	0.88^{a}	0.81^{a}	0.30	***	n.s.
C18:3*n*-3	15.2^{a}	21.4^{b}	22.2^{by}	21.2^{by}	11.6	16.9	13.7^{x}	14.7^{x}	0.98	**	***
C18:2*c*9 *t*11	1.70	1.15	1.39	1.51	1.67	1.29	1.11	1.33	0.17	n.s.	n.s.
C20:2	1.23^{b}	0.36^{a}	0.63^{a}	0.85^{a}	1.11^{b}	0.45^{a}	0.43^{a}	0.36^{a}	0.09	***	n.s.
C22:0	2.04	2.07	1.70	1.72	1.91	1.99	0.88	1.02	0.35	n.s.	n.s.
C20:3*n*-6	5.39^{y}	4.87	5.71^{y}	5.98^{y}	3.33^{x}	3.55	3.31^{x}	3.38^{x}	0.28	n.s.	***
C20:3*n*-3	0.39	0.21	0.40	0.26	0.23	0.30	0.17	0.10	0.06	n.s.	n.s.
C20:4*n*-6	21.7^{y}	22.9^{y}	25.7^{y}	25.3^{y}	12.3^{x}	15.6^{x}	13.7^{x}	14.7^{x}	1.14	n.s.	***
C20:4*n*-3	0.27	0.61	0.38	0.20	0.09	0.37	0.32	0.37	0.11	n.s.	n.s.
C22:2	1.62	1.89	2.58	2.72	0.93	1.45	1.15	1.52	0.29	n.s.	**
C24:0	0.34	0.16	0.17	0.28	0.25	0.24	0.26	0.20	0.05	n.s.	n.s.
C20:5*n*-3	5.26^{a}	9.32^{b}	9.32^{by}	9.11^{by}	3.35	6.11	4.62^{x}	4.97^{x}	0.56	**	***
C22:5*n*-3	10.7^{y}	12.1	13.1^{y}	12.8^{y}	5.34^{x}	8.87	6.78^{x}	7.33^{x}	0.61	*	***
C22:6*n*-3	0.79^{a}	1.22^{ab}	1.56^{by}	1.53^{by}	0.31	0.82	0.77^{x}	0.79^{x}	0.10	**	***
Others	4.37	1.81	2.94	3.90	4.58	2.71	2.64	2.79	0.59	n.s.	n.s.
Total	2158^{b}	638^{a}	869^{a}	880^{a}	2255^{b}	644^{a}	753^{a}	766^{a}	130	***	n.s.
SFA	1039^{b}	294^{a}	403^{a}	416^{a}	1102^{b}	314^{a}	381^{a}	390^{a}	66	***	n.s.
MUFA	947^{b}	201^{a}	300^{a}	291^{a}	1026^{b}	220^{a}	272^{a}	263^{a}	61	***	n.s.
PUFA	168^{y}	141	163^{y}	169^{y}	122^{x}	108	98^{x}	111^{x}	7.00	n.s.	***
PUFA : SFA	0.18^{a}	0.58^{b}	0.51^{b}	0.52^{b}	0.12^{a}	0.41^{b}	0.32^{b}	0.33^{b}	0.04	***	***
n-6 PUFA	135^{b}	96^{a}	116^{aby}	124^{aby}	101	75	72^{x}	83^{x}	6.00	**	***
n-3 PUFA	32.6^{a}	44.9^{b}	47.0^{by}	45.1^{by}	20.9^{a}	33.3^{b}	26.4^{abx}	28.3^{abx}	2.01	**	***
n-6 *: n*-3	4.27^{b}	2.15^{a}	2.50^{a}	3.00^{a}	5.00^{b}	2.32^{a}	2.79^{a}	3.08^{a}		***	n.s.
HP-PUFA	60.1^{y}	73.0^{y}	78.9^{y}	76.9^{y}	36.9^{x}	52.8^{x}	43.6^{x}	46.6^{x}	3.09	*	***
α-Tocopherol/PUFA*10^{-4}	15^{y}	23^{y}	18^{y}	14^{y}	3.3^{ax}	8.6^{bx}	6.3^{abx}	6.7^{bx}	11	***	***
α-Tocopherol/HP-PUFA	39^{y}	44^{y}	38^{y}	30^{y}	11^{x}	17^{x}	13^{x}	16^{x}	20	n.s.	***
Atherogenicity index	0.75^{b}	0.49^{a}	0.56^{a}	0.57^{a}	0.75^{b}	0.55^{a}	0.66^{ab}	0.67^{ab}	0.03	***	*
Thrombogenic index	1.54^{b}	0.93^{a}	1.04^{a}	1.08^{a}	1.68^{b}	1.17^{a}	1.40^{ab}	1.41^{ab}	0.06	***	***

C = concentrate; P = grass only; PC25 = grass with 25% concentrate DM; PC50 = grass with 50% concentrate DM; SFA = saturated fatty acids; MUFA = monounsaturated fatty acids; PUFA = polyunsaturated fatty acids; HP-PUFA = highly peroxidisable PUFA.

[1]Samples were stored in modified atmosphere (O_2 : CO_2; 80 : 20) and subjected to simulated retail display (4°C, 1000 lux for 12 h out of 24 h) for 14 days.

HP-PUFA: Calculated as the sum of PUFA with three or more double bonds.

α-Tocopherol : PUFA: ratio between the concentration of α-tocopherol and the concentration of PUFA, both expressed in mg/g of muscle.

α-Tocopherol : HP-PUFA: ratio between the concentration of α-tocopherol and the concentration of highly peroxidisable PUFA, both expressed in mg/g of muscle.

Atherogenic index: $(C12:0 + 4 \times C14:0 + C16:0)/(\sum PUFA + \sum MUFA)$.

Thrombogenic index: $(C14:0 + C16:0 + C18:0)/(0.5 \times \sum MUFA + 0.5 \times n\text{-}6 + 3 \times n\text{-}3 + n\text{-}3/n\text{-}6)$.

[a,b,c]Treatment means within rows, on the same storage day, assigned different superscripts differ significantly, $P < 0.05$.

[x,y]Treatment means within rows on different days of storage, assigned different superscripts differ significantly, $P < 0.05$.

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

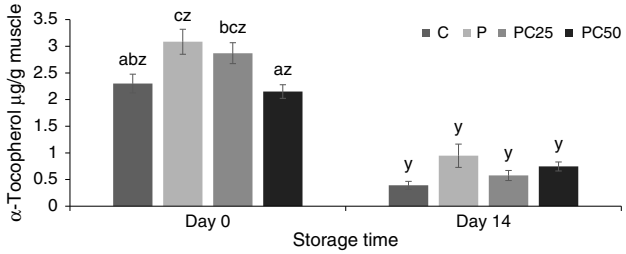

Figure 1 The α-tocopherol concentration in *M. longissimus thoracis et lumborum* of late-maturing bulls from four production systems (C: rolled barley concentrate, P: grass only, PC25: grass with 25% concentrate DM and PC50: grass with 50% concentrate DM) stored in modified atmosphere (O_2 : CO_2; 80 : 20) and subjected to simulated retail display (4°C, 1000 lux for 12 h out of 24 h) for 0 and 14 days. [a,b,c]Within storage time, least square means assigned different superscripts differ ($P < 0.05$) between production systems. [y,z]Within production system, least square means assigned different superscripts differ ($P < 0.05$) due to storage time.

There was a tendency for the proportion of C18:2*n*-6*t* to increase ($P = 0.07$) while that of C20:2 tended to decrease ($P = 0.07$) with increase in storage time. Storage time did not have an effect ($P > 0.05$) on the proportions of C10:0, C12:0, C14:0, C14:1, C16:1, C17:0, C17:1, C18:1*n*-7, C20:0, C20:1*n*-9, C18:2*c*9 *t*11, C22:0, C20:3*n*-3, C20:4*n*-3 and C24:0. The proportion of MUFA was higher ($P = 0.02$) on day 14 compared to day 0. The proportions of PUFA, *n*-6 PUFA and *n*-3 PUFA were lower ($P < 0.001$) on day 14 relative to day 0.

Muscle α-tocopherol concentration

The main effects of PS and storage time on muscle α-tocopherol concentration are shown in Figure 1. There was an interaction ($P = 0.05$) between PS and storage time for α-tocopherol concentration such that on day 0 muscle from P and PC25 bulls, which did not differ ($P > 0.05$), had higher α-tocopherol concentration than PC50 but they were not significantly different on day 14. Muscle from P bulls had a 1.3-fold higher ($P < 0.001$) α-tocopherol concentration compared to C bulls. α-Tocopherol concentration in muscle from C bulls was similar to that in muscle from PC50 bulls ($P > 0.05$). α-Tocopherol concentration did not differ between PS on day 14. α-Tocopherol concentration declined as TBARS values increased (Figure 2) such that at day 14 only about 21% of day 0 α-tocopherol remained in muscle.

There was an interaction between PS and storage day ($P < 0.001$) for α-tocopherol : PUFA ratio in muscle such that although there were no effects of PS on day 0, there was a lower ratio in C compared to P and PC50 on day 14 (Table 4). The muscle α-tocopherol : PUFA ratio was higher ($P < 0.001$) in P and PC25 than in C, while PC50 had a lower ratio compared to P but had a similar ratio to C and PC25. There was no effect of PS on muscle α-tocopherol : HP-PUFA ratio. The muscle α-tocopherol : PUFA and α-tocopherol : HP-PUFA ratios were both lower on day 14 compared to day 0 with significant decreases across all PS.

Colour

The effect of PS on colour (lightness and redness) is shown in Table 5. There was no interaction ($P = 0.82$) between PS and storage day for lightness. There was an overall effect of PS on lightness whereby muscle from C and P bulls, which did not differ, was significantly darker than muscle from PC50 bulls ($P = 0.02$). Lightness of muscle from PC25 was similar ($P > 0.05$) to lightness of muscle from P, PC50 and C bulls. For redness there was an interaction ($P = 0.01$) between PS and storage day whereby there were significant differences ($P < 0.001$) between C and P on day 10 while there were no differences on the other storage days. Lightness on day 0 and 3, which did not differ ($P > 0.05$), was lower ($P < 0.001$) than on days 7, 10 and 14, which also did not differ ($P > 0.05$). There was an initial increase ($P < 0.001$) in redness between day 0 and day 3 and then redness decreased with an increase in storage time.

Lipid oxidation

Production system and storage time had a significant effect on TBARS values ($P < 0.001$) (Figure 2). There was an interaction between PS and storage time ($P = 0.009$) such that while there were no PS effects ($P > 0.05$) on TBARS values on days 0, 3 and 7, on day 10 muscle from P bulls had lower TBARS values than muscle from C ($P = 0.02$) and PC50 ($P < 0.001$) bulls and on day 14, muscle from P bulls had lower ($P < 0.001$) TBARS values than muscle from C and PC25 bulls. There were no differences in TBARS values ($P > 0.05$) between muscle from PC25 and PC50 and C. As the days of storage progressed TBARS values in muscle increased ($P < 0.001$).

Correlations between α-tocopherol concentration, fatty acid profile and 2-thiobarbituric acid-reactive substances values in muscle

Pearson correlations between α-tocopherol concentration, FA profile and TBARS values in muscle are shown in Table 6. There were negative correlations between TBARS values and α-tocopherol concentration ($r = -0.26$, $P < 0.01$), α-tocopherol : PUFA ($r = -30$, $P < 0.01$), HP-PUFA ($r = -0.25$, $P < 0.05$), *n*-3 PUFA ($r = -0.28$, $P < 0.05$), PUFA : SFA ratio ($r = -0.35$, $P < 0.001$) and positive correlations between muscle TBARS values and the *n*-6 : *n*-3 ratio ($r = 0.47$, $P < 0.001$).

Discussion

Feed fatty acid composition and α-tocopherol concentration

The feed FA composition data confirm that high energy grain-based diets are sources of mainly SFA and MUFA, while grass

Table 5 *Colour (lightness 'L' and redness 'a') of* M. longissimus thoracis et lumborum *muscle of late-maturing bulls from four production systems (PS)*

		Production system					*P*-values		
	Day[1]	C	P	PC25	PC50	SEM	PS	Day	PS*Day
L	0	36.4	35.0^{v}	35.6	36.9	0.24	*	***	n.s
	3	35.4	35.7^{vw}	35.9	36.8				
	7	36.6	37.0^{vw}	37.4	37.9				
	10	37.2	37.6^{w}	37.4	38.5				
	14	37.5	37.7^{w}	37.1	37.6				
a	0	11.3^{x}	11.1^{x}	11.5^{x}	11.7^{x}	0.20	n.s	***	*
	3	15.6^{z}	14.1^{y}	14.3^{y}	14.2^{z}				
	7	13.3^{y}	13.1^{xy}	13.6^{xy}	13.6^{xy}				
	10	6.39^{aw}	8.57^{bw}	7.99^{abw}	8.29^{abw}				
	14	4.62^{v}	6.08^{v}	5.75^{v}	5.80^{v}				

C = concentrate; P = grass only; PC25 = grass with 25% concentrate DM; PC50 = grass with 50% concentrate DM.
[1]Samples were stored in modified atmosphere (O_2 : CO_2; 80 : 20) and subjected to simulated retail display (4°C, 1000 lux for 12 h out of 24 h) for up to 14 days.
[a,b]Least square means within rows assigned different superscripts differ significantly ($P < 0.05$).
[v,w,x,y,z]Least square means within columns assigned different superscripts differ significantly ($P < 0.05$).
*$P < 0.05$; ***$P < 0.001$.

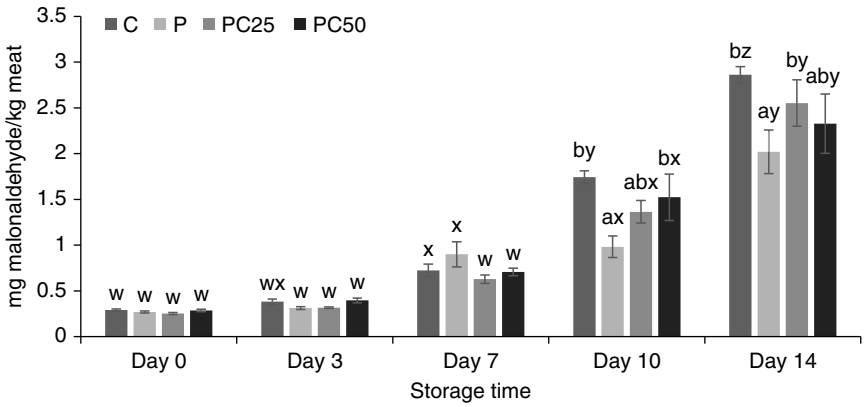

Figure 2 Lipid oxidation (2-thiobarbituric acid-reactive substances values) in *M. longissimus thoracis et lumborum* of late-maturing bulls from four production systems (C: concentrate, P: grass only, PC25: grass with 25% concentrate DM and PC50: grass with 50% concentrate DM) stored in modified atmosphere (O_2 : CO_2; 80 : 20) and subjected to simulated retail display (4°C, 1000 lux for 12 h out of 24 h) for 0, 3, 7, 10 and 14 days. [a,b]Within storage time, least square means assigned different superscripts differ ($P < 0.05$) between production systems. [w,x,y,z]Within production system, least square means assigned different superscripts differ ($P < 0.05$) due to storage time.

or forage-based diets are sources of PUFA, particularly *n*-3 PUFA. The α-tocopherol concentration of grass, silage and rolled barley concentrate in this study was similar to the values reported in the study by Röhrle *et al.* (2011). The threefold higher α-tocopherol concentration in grass silage compared to grass may be a consequence of different sampling times or growth stage for the grass *v.* the grass for ensiling. It may also be a result of α-tocopherol being more stable in an acidic environment devoid of oxygen and light as well as the transformation of tocotrienols to tocopherols in the moist and airtight conditions of a silo (Müller *et al.*, 2007). Difficulties in obtaining representative samples, settling of minor ingredients or degradation during storage of concentrates could explain why the value for α-tocopherol in the concentrate was less than 10% of the predicted value in the feed.

Proximate composition

The higher IMF concentration in concentrate finished beef observed in this study is comparable to observations in other studies (Nuernberg *et al.*, 2005, Leheska *et al.*, 2008) and is attributable to the higher intake of energy and protein from concentrates in contrast to pasture. Total DMI for P, PC25 and PC50 bulls was 84%, 92% and 89% that of the C bulls, respectively, and the estimated daily energy intakes for P, PC25 and PC50 bulls were 66, 75 and 76% of that of C bulls, respectively (Lenehan, 2016). As expected, average daily gains (kg) of 0.61, 0.57, 0.80 and 1.08, leading to carcass weights (kg) of 367, 367, 387 and 406 for P, PC25, PC50 and C, respectively, were recorded. In addition, the higher IMF concentration in concentrate finished beef compared to those finished on pasture could be due to higher energy expenditure during exercise in pasture finished cattle (Dunne *et al.*, 2005b). It is noteworthy that the C bulls

Table 6 *Correlations between α-tocopherol concentration, fatty acid profile and TBARS values in* M. longissimus thoracis et lumborum of late-maturing bulls

	α-T : PUFA	α-T : HP-PUFA	PUFA	HP-PUFA	*n*-6 PUFA	*n*-3 PUFA	TBARS	PUFA : SFA	*n*-6 : *n*-3
α-T	0.88***	0.89***	0.52***	0.68***	0.40***	0.66***	−0.26**	0.21*	−0.32**
α-T : PUFA		0.97***	0.13	0.37***	0.01	0.39***	−0.30**	0.30**	−0.41***
α-T : HP-PUFA			0.21*	0.35***	0.13	0.33***	−0.18	0.22*	−0.25**
PUFA				0.84***	0.97***	0.75***	0.04	−0.13	0.13
HP-PUFA					0.70***	0.96***	−0.25*	0.11	−0.36***
n-6 PUFA						0.57***	0.16	−0.21*	0.35***
n-3 PUFA							−0.28**	0.12	−0.51***
TBARS								−0.35***	0.47***
PUFA : SFA									−0.38***

α-T = α-tocopherol concentration; α-T : PUFA = α-tocopherol concentration to polyunsaturated fatty acid ratio; α-T : HP-PUFA = α-tocopherol concentration to highly peroxidisable polyunsaturated fatty acid ratio; PUFA = polyunsaturated fatty acids; HP-PUFA = highly peroxidisable polyunsaturated fatty acids; *n*-6 PUFA = *n*-6 polyunsaturated fatty acids; *n*-3 PUFA = *n*-3 polyunsaturated fatty acids; TBARS = 2-thiobarbituric acid-reactive substances; PUFA : SFA = polyunsaturated fatty acids to saturated fatty acids ratio.
*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

had more than fourfold higher IMF than the pasture-based P, PC25 and PC50 bulls. The IMF also reflects the carcass fat scores, which were at least 1.5-fold higher for the C bulls (Lenehan, 2016). The IMF reported in the current study is lower than that reported in other studies (Nuernberg *et al.*, 2005, Mezgebo *et al.*, 2017b). However, comparable muscle IMF values were reported by Mezgebo *et al.* (2017a) in late-maturing type bulls similar to those used in this study.

Muscle fatty acids

Differences in the intramuscular FA composition reflected feed FA composition. Similar to other studies, the inclusion of grass or forage in the diet led to decreases in the concentrations of the C14:0, C14:1 C15:0, C16:0, C16:1, C17:0, C18:0, C18:1*n*-9*c*, C18:1*n*-7, C20:1*n*-9 SFA, C20:2, total FA, SFA and MUFA in the muscle of P, PC25 and PC50 bulls (Mezgebo *et al.*, 2017b). Higher muscle concentrations of C18:3*n*-3, C20:5*n*-3, C22:6*n*-3, total *n*-3 PUFA and high HP-PUFA in P, PC25 and PC50 were expected since forage diets tend to increase *n*-3 PUFA (Warren *et al.*, 2008). With regard to nutritional indices a higher PUFA : SFA ratio and lower *n*-6 : *n*-3 PUFA ratio in P, PC25 and PC50 compared to C agrees with other studies (Enser *et al.*, 1998, French *et al.*, 2000). Considering European Food Safety Authority Regulations for Health Claims (European Commission, 2012), none of PS produced meat which could be labelled as a source of *n*-3 PUFA. A claim that a food is a source of *n*-3 PUFA can be made on foods containing at least 0.3 g C18:3*n*-3/100 g and per 100 kcal, or at least 40 mg of the sum of C20:5*n*-3 and C22:6*n*-3 per 100 g and per 100 kcal. The muscle from P, PC25 and PC50 had only 25% of the sum of C20:5*n*-3 and C22:6*n*-3 required to be considered a source of *n*-3 PUFA, whereas muscle from C bulls had only 15% of the requirement. The PUFA : SFA ratios in the pasture-based groups were higher than in other studies (Enser *et al.*, 1998, French et al., 2000), and this is most likely due to low IMF which increases the significance of the phospholipid portion of FA in the total lipid concentration (Warren *et al.*, 2008). The higher HP-PUFA in muscle of P, PC25 and PC50 compared to C was in agreement with Luciano *et al.* (2011). The AI and TI data showed that beef produced from pasture fed bulls, including those receiving concentrate supplementation at up to 50% of DM, had a healthier profile than beef produced from concentrate fed bulls, but the extent of the differences was diminished with storage in MAP most likely due to oxidation of unsaturated fatty acids. Overall, despite supplementation of pasture with concentrate up to 50% (DM basis) the effect of grass feeding on the FA profile was still evident as the only observed difference in muscle FA profile among the grass-based bulls was a higher *n*-6 : *n*-3 ratio in PC50 in relation to P.

Comparison of FA composition in muscle from different production systems using absolute values may be confounded by the effects of IMF since the neutral lipid proportion of fat increases with increase in fatness, while the phospholipid portion remains fairly constant (Warren *et al.*, 2008). With an increase in neutral lipid concentration the proportions of SFA and MUFA increase (Enser *et al.*, 1998, Warren *et al.*, 2008). Use of proportions rather than absolute values allows a comparison of treatments at an 'equal' total lipid concentration, which is especially useful in this study where differences in IMF concentration are large. The higher muscle proportion of C18:0 in P, PC25 and PC50 compared to C (Supplementary Material S1) is consistent with observations in other studies (Warren *et al.*, 2008; Daley *et al.*, 2010). It is desirable to have low amounts of SFA such as C16:0 since they are atherogenic and hypercholesterolemic; however, C18:0 is believed to be neutral in its effect (Crupkin and Zambelli, 2008). The higher proportion of C18:0 and lower proportion MUFA in P, PC25 and PC50 in relation to C in this study contrasts with findings of French *et al.* (2000) where levels were the same for concentrate-fed and grass-fed animals. The higher MUFA proportion in muscle from C bulls compared to the P, P50 and P25 confirms that MUFA increases with an increase in fatness. The difference between this study and French *et al.* (2000) is that in the latter study

there were no differences in IMF, and IMF was higher with all treatments having more than 3% IMF. The higher proportions of PUFA in P, PC25 and PC50 compared to C is similar to observations in other studies where grass-fed beef and concentrate-fed beef were compared (Leheska *et al.*, 2008). Grass feeding tends to increase the proportion of PUFA, especially *n*-3 PUFA. Long chain *n*−6 and *n*−3 PUFA are synthesised from C18:2*n*−6 and C18:3*n*−3, respectively (Warren *et al.*, 2008), explaining the higher proportions of C20:5*n*-3 and C22:5*n*-3 observed in P, PC25 and PC50 compared to C. Expression of muscle FA profile on a proportion basis showed some differences which were not observed when expressed as a concentration. Supplementation of pasture with concentrate (PC25, PC50) resulted in lower muscle proportions of C18:3*n*-3, C22:0, C22:5*n*-3, *n*-3 PUFA and HP-PUFA relative to P. Similarly, French *et al.* (2000) showed higher *n*-3 PUFA proportion in steers on pasture *v.* those finished on supplemented pasture.

The susceptibility of FA to oxidation increases, while time to initiation of lipid oxidation decreases, as the number of double bonds increases (Belitz *et al.*, 2009). Therefore, the decrease in PUFA with three or more double bonds (C18:3*n*-6, C18:3*n*-3, C20:3*n*-6, C20:4*n*-6, C20:5*n*-3, C22:5*n*-3, C22:6*n*-3, *n*-3 PUFA and HP-PUFA) while the concentrations of SFA remained unchanged was not unexpected. The decrease in the PUFA : SFA ratios with storage in muscle of C, PC25 and PC50 bulls, with no decrease in the PUFA : SFA ratio in muscle of P bulls, indicates greater antioxidant protection of muscle HP-PUFA in P.

Muscle α-tocopherol concentration

The lower α-tocopherol concentration in C compared to P is consistent with observations in several studies (Liu *et al.*, 1995, Descalzo *et al.*, 2005, Luciano *et al.*, 2011). The average daily DMI of grass for the P, PC25 and PC50 bulls during the finishing phase was estimated to be 8.7, 7.1 and 4.4 kg DM, respectively, while the average daily DMI of silage was 1.5 kg for the C bulls. Concentrate DMI was 2.4, 4.8 and 10.3 kg for PC25, PC50 and C bulls, respectively (Lenehan, 2016). Using the average daily DM intake values and the measured concentration of α-tocopherol in the feedstuffs (Table 2), the intake of α-tocopherol of the bulls finished on P, PC25, PC50 and C was estimated to be 183, 187, 169 and 249 mg/animal per day during the 120 days of finishing. However, while the concentrate finished bulls were fed α-tocopherol acetate supplemented feed such that α-tocopherol intakes were expected to be higher for C compared to P, PC25 and PC50, the bioavailability of *all-rac* α-tocopherol is lower than that of natural α-tocopherol in grass and forages. Lower bioavailability is due to preferential tissue deposition of the *RRR* α-tocopherol isomer over the 2S isomers which constitute 50% of the synthetic α-tocopherol that is included in the concentrate rations (Weiss *et al.*, 2009). Alternatively, higher α-tocopherol in pasture finished groups could be due to regeneration and protection of α-tocopherol by other antioxidants such as ascorbic acid and β-carotene (Niki, 2014). The higher α-tocopherol : PUFA and α-tocopherol : HP-PUFA ratios in muscle from P bulls may explain the higher resistance to lipid oxidation compared to other treatments (Luciano *et al.*, 2011).

α-Tocopherol delays lipid oxidation and metmyoglobin formation by quenching radicals through donation of a hydrogen atom (Niki, 2014); therefore α-tocopherol would be expected to decrease as TBARS values increased. Insani *et al.* (2008) reported a 41% and 57% decrease in α-tocopherol concentration after 9 days of simulated retail display (4°C, 1900 lux) of unaged meat samples from pasture-fed and grain-fed cattle, respectively. Similarly, Descalzo *et al.* (2008) observed a dramatic decline of α-tocopherol concentration during ageing of buffalo meat in vacuum packs at 4°C with levels averaging 4.22 μg/g in fresh meat and 1.37 μg/g after 15 days of ageing and 0.122 μg/g after 25 days of ageing.

Colour and lipid oxidation

In contrast to several authors who observed darker meat from pasture compared to a concentrate production system (Vestergaard *et al.*, 2000, Lanari *et al.*, 2002, Yang *et al.*, 2002) the P group had similar lightness to C. Grazing animals have higher muscle myoglobin concentrations and are also more likely to be stressed at slaughter as they may not be accustomed to handling, leading to high muscle pH and consequently dark cutting meat (Vestergaard *et al.*, 2000). There was no evidence of preslaughter stress as all muscles had pH within the normal range of 5.4 to 5.8 (Tarrant, 1989), and none of the carcasses were declared as dark cutting in the abattoir, which could explain the similar lightness between P and C.

Consumer perception of redness at the point of sale determines the marketability of fresh meat, hence the importance of examining the effect of PS on colour stability during retail display. Oxymyoglobin oxidation is related to lipid oxidation-induced depletion of dissolved oxygen and therefore α-tocopherol extends colour shelf life (Monahan *et al.*, 2005). In this study muscle from P showed higher resistance to colour deterioration as redness was higher than in muscle from C bulls on day 10 of storage. Similar results were reported by Insani *et al.* (2008) where redness in *M. psoas major* from pasture-finished beef was higher than in maize grain-finished samples at day 7 of storage. On the other hand, Lanari *et al.* (2002) found similar redness values during storage days 6 to 14 in unaged *gluteus medius* (**GM**) from Hereford sired steers finished on pasture or sorghum-based concentrate supplemented with 2500 IU/head per day of *all-rac* α-tocopherol acetate. The threshold amount of α-tocopherol required to maintain colour stability is reported to be 3.0 to 3.5 μg/g (Liu *et al.*, 1995); none of the treatments in this study attained a muscle α-tocopherol concentration of > 3.0 μg/g except P. The higher redness value of muscle from P bulls *v.* C bulls on day 10 of refrigerated storage may be due to the lower muscle α-tocopherol concentration of the latter.

The observation that TBARS values were higher in C compared to P on days 10 and 14 is similar to observations

in other studies (Descalzo *et al.*, 2005, Insani *et al.*, 2008, Fruet *et al.*, 2018). In contrast Luciano *et al.* (2011) found no differences and Yang *et al.* (2002) found lower TBARS values in beef from animals fed α-tocopherol fortified concentrate when pasture- and concentrate-fed steers were compared. Higher muscle α-tocopherol concentration and higher α-tocopherol : PUFA ratio in P compared to C resulted in higher resistance to lipid oxidation in P compared to C. Resistance to lipid oxidation could also be due to higher levels of other antioxidants (not measured in this study) in P such as β-carotene, vitamin C, flavonoids and polyphenols as well as endogenous antioxidants such as superoxide dismutase and catalase (Daley *et al.*, 2010). Similar TBARS values on days 10 and 14 between PC25, PC50 and C may be due to a lower α-tocopherol : PUFA ratio in the supplemented pasture diets.

Correlations between α-tocopherol concentration, fatty acid profile and 2-thiobarbituric acid-reactive substances values in muscle

The negative correlation between TBARS values and α-tocopherol concentration indicates that lipid oxidation increases as α-tocopherol concentration decreases (Table 6) which has previously been shown by Ponnampalam *et al.* (2014). The absence of a relationship between PUFA and TBARS values indicates that PUFA may not be the main driver of lipid oxidation or is perhaps a reflection of the similar amounts of PUFA in all PS. On the other hand, the negative correlation between HP-PUFA and TBARS may be due to the fact that samples with higher HP-PUFA also had higher α-tocopherol concentration. This is supported by Ponnampalam *et al.* (2014) who found that α-tocopherol concentration explained most of the effect of diet on lipid oxidation. The negative correlations between TBARS values and the α-tocopherol : PUFA and α-tocopherol : HP-PUFA ratios despite the lack of a relationship between PUFA and TBARS values may indicate that antioxidant concentration and the ratio of antioxidant to prooxidant components may be more important than the amount of PUFA considered separately.

Conclusion

Finishing of late-maturing bulls on grass with or without supplementation with cereal concentrate results in beef with a low IMF concentration and higher PUFA (including long chain highly peroxidisable PUFA) proportion but higher lipid stability under retail storage conditions compared to concentrate finishing. Pasture finishing even when concentrate constitutes 50% of dietary DMI results in a more nutritionally favourable fatty acid profile compared to concentrate finishing. Feeding up to 50% concentrates at pasture preserves the *n*-3 PUFA concentration in muscle without any decrease in oxidative stability.

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F. J. Monahan 0000-0002-0938-6435

Declaration of interest

The authors confirm that there are no conflicts of interest.

Ethics statement

All animal procedures used in this study were conducted under experimental license from the Health Products Regulatory Authority (HPRA) in accordance with the European Union *Protection of Animals used for Scientific Purposes Regulations 2012* (S.I. No. 543 of 2012).

Software and data repository resources

Data are not deposited in an official repository.

Supplementary material

To view supplementary material for this article, please visit https://doi.org/10.1017/S1751731119002313

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