Published September 7, 2017

Meat and Muscle Biology™

Palatability of Beef from Cattle Fed Extruded Flaxseed before Hay or Mixed with Hay

N. Prieto,* M. E. R. Dugan, I. L. Larsen, P. Vahmani, and J. L. Aalhus

Lacombe Research and Development Centre, Agriculture and Agri-Food Canada, Lacombe, Alberta, T4L 1W1, Canada
*Corresponding author. E-mail: nuria.prietobenavides@agr.gc.ca (N. Prieto)

Abstract: This study was undertaken to evaluate the eating quality of *longissimus thoracis* steaks and hamburgers (80/20 *gluteus medius* to perirenal fat) with enhanced profiles of potentially healthy fatty acids. The profile of health favorable fatty acids (n-3, vaccenic and rumenic) was improved in beef by feeding co-extruded flaxseed (flaxseed, peas, and alfalfa) and alfalfa-grass hay as a total mixed ration (TMR), and further enhanced by feeding co-extruded flaxseed before alfalfa-grass hay (Non-TMR). Compared to TMR, feeding steers the Non-TMR resulted in tougher steaks ($P < 0.05$) with lower beef flavor ($P < 0.01$) and greater off-flavor ($P < 0.01$) intensity to an extent that might be detectable by consumers. High levels of *trans*-monounsaturated fatty acids, mainly of vaccenic acid, were associated with a fishy off-flavor, although actual changes in flavor may relate to correlated combined effects of conjugated fatty acids, atypical dienoic acids and α-linolenic acid. Diet had no significant effect on sensory attributes of hamburgers, but when panelists described off-flavors, they noted more 'other' off-flavors ($P < 0.05$) with fishy and stale/cardboard notes being more prominant in Non-TMR hamburgers. Overall, beef samples with threshold levels of vaccenic acid over 6.12% of total fatty acids resulting from feeding flaxseed products, while of potentially greater health benefit, may pose challenges in terms of eating quality. Areas worthy of further investigation to ensure acceptable eating quality might be the influence of ageing on antioxidant capacity in beef with enhanced fatty acid profiles, and the potential use of protective packaging to limit deterioration.

Keywords: beef, eating quality, flavor, healthful fatty acids
Meat and Muscle Biology 1:157–168 (2017) **doi:10.22175/mmb2017.04.0026**
Submitted 27 Apr. 2017 Accepted 21 July 2017

Introduction

When consumers purchase beef, quality and price are considered key factors (Realini et al., 2014). However, an increasing number of informed consumers from medium to high incomes are also factoring in the health implications of beef consumption to their meat purchases (Kallas et al., 2014; Realini et al., 2014). Because of this, there have been many attempts to increase n-3 fatty acids in beef due to their health benefits and their presence in ruminant animals (Benjamin and Spener, 2009; Field et al., 2009).

In cattle, microbial lipases bind to dietary lipids present in the rumen and release free fatty acids such as polyunsaturated fatty acids (PUFA), which are toxic to rumen microbes (Jenkins et al., 2008). To reduce the toxicity, PUFA are then biohydrogenated by the rumen microbes, producing saturated fatty acid (SFA) of lower toxicity, particularly 18:0. The residual PUFA biohydrogenation intermediates (PUFA-BHI) originated during this process bypass the rumen, and are absorbed from the lower gut and incorporated into tissues, including muscle (Scollan et al., 2014). Hence, beef can also be a source of PUFA-BHI, including *t*11–18:1 (vaccenic acid) and *c*9,*t*11–18:2 (rumenic acid), which may have beneficial health effects related to cancer, inflammatory

This project was funded by the Alberta Meat and Livestock Agency (2015P006R). The gift of LinProTM-R from O&T Farms (Regina, SK, Canada) is gratefully acknowledged. The authors also thank the Lacombe Research and Development Centre operational, processing and technical staff for their dedication and expert assistance.

diseases, type II diabetes and post-menopausal osteoporosis (Kim et al., 2016; Field et al., 2009). Hence, in addition to n-3 fatty acids, considerable efforts have been made to try and increase rumenic and vaccenic acids in beef, but often with limited success (Vahmani et al., 2015; Nassu et al., 2011; Kronberg et al., 2006).

In a recent study, however, Vahmani et al. (2017) may have found a key to increasing n-3 fatty acids, and rumenic and vaccenic acids in beef. These authors hypothesized that feeding extruded flaxseed prior to hay in a non-total mixed ration (non-TMR) would reduce the extent of biohydrogenation in the rumen, and increase the concentrations of 18:3n-3 (α-linolenic acid) and its BHI in beef. When Vahmani et al. (2017) fed extruded flaxseed prior to feeding hay in a Non-TMR, it significantly increased α-linolenic, rumenic and vaccenic acids, and a number of other α-linolenic-BHI in beef compared to feeding the same ingredients in a TMR. Specifically, feeding the Non-TMR enriched *longissimus thoracis* (LT) and hamburger with α-linolenic acid (+12% and +17%, respectively), vaccenic acid (+49% and +39%, respectively), rumenic acid (+33% and +47%, respectively), conjugated linolenic acid (CLnA; +49% and +67%, respectively), *t*11,*c*13–18:2 (+93.7% and +102%, respectively), *t*11,*t*15–18:2 (+67% and +46%, respectively), and *t*11,*c*15–18:2 (+40% and +33%, respectively).

However, beef with enhanced healthful fatty acids needs to be tested in terms of sensory quality because the fatty acid composition (both proportions and amounts) in intramuscular fat have been shown to have a large influence on beef quality (Wood et al., 2004). Unsaturated fatty acids are prone to rapid oxidation, especially fatty acids with 2 or more double bonds. This increases the rate of rancidity development and color deterioration of meat, and can affect the shelf life of meat (Wood et al., 2004). Additionally, oxidation of unsaturated fatty acids results in an important source of volatiles compounds that can affect flavor development during cooking (Mottram, 1998). Individual fatty acids have a wide range of melting points, and therefore different compositions can change the fat firmness, especially in subcutaneous and intermuscular fat deposits, but also the intramuscular fat. Changes to the firmness of intramuscular fat can then affect meat tenderness (Wood, 1990). In fact, Elmore et al. (2002) and Miller (2001) indicated there is a desire to increase the PUFA in meat for human health benefits. However, when PUFA are too abundant, the oxidation developed can result in a shorter shelf life and negative flavors or off-flavors in cooked meat.

Hence, the aim of this study was to evaluate the eating quality of beef (steaks and hamburgers) with enhanced profiles of potentially healthy fatty acids (n-3, vaccenic and rumenic) from steers fed extruded flaxseed and ground hay either together as a TMR or sequentially (Non-TMR).

Material and Methods

Animals and diets

Forty-eight Angus cross steers (325.2 ± 15.9 kg) were used in this study at the Lacombe Research and Development Centre (Lacombe RDC, Alberta, Canada). Steers were cared for according to Canadian Council on Animal Care guidelines and the study was approved by the Lacombe RDC Animal Care Committee (CCAC, 2009). Live weights 1 d prior to the start of the trial were used to stratify steers before randomizing to one of 6 pens (8 steers per pen). Pens were 110 m^2 with concrete surfaces and 9 m of concrete feed bunk per pen. Pens of 8 steers were randomized to either a TMR or a Non-TMR treatment with 3 pens per treatment. The TMR included linPRO-R [50% co-extruded linseed, 40% field peas, and 10% alfalfa; 25% of dry matter (DM); O&T Farms, Regina, SK], tub ground hay (alfalfa-grass mix; 74.75% of diet DM) and a vitamin supplement (0.25% of DM providing 2,200, 440, and 135 IU of vitamin A, D3, and E per kg of diet), which were mixed together and fed daily at 8:30 h. The Non-TMR treatment included the same ingredients with the same proportions as the TMR treatment, but linPRO-R and vitamin supplement were fed at 8:30 h followed by tub ground hay at 11:30 h. Steers in each pen were group fed to appetite with feed supply targeted to ensure all feed being consumed by 24 h. For the Non-TMR group, all of the linPRO-R was consumed before the hay was fed. Steers had free access to fresh, clean water. Steers also had ad libitum access to trace mineral salt (96.5% NaCl, 4,000 mg × kg^{-1} Zn; 1,600 mg × kg^{-1} Fe; 1,200 mg × kg^{-1} Mn; 330 mg × kg^{-1} Cu; 100 mg × kg^{-1} I; 40 mg × kg^{-1} Co; Compass Minerals, Mississauga, ON, Canada).

Slaughter procedure and sample collection

Steers were slaughtered over a 1 mo period, with the 2 steers with the greatest backfat thickness from each pen slaughtered per week with an average of 242 ± 10.5 d on experimental diets. Steers were slaughtered at the federal inspected Lacombe RDC research abbatoir. At slaughter, steers were stunned, exsanguinated and dressed in a commercial manner. During evisceration, perirenal fat was collected as the primary fat source and

stored at 2°C for 13 d to manufacture ground beef in an 80/20 lean to fat ratio. Following carcass splitting, carcass sides were chilled for 72 h in a cooler set at 2°C, with an average wind speed of 1.4 m × s^{-1}. At 72 h post-slaughter, the left LT was dissected from the carcass and a 2.5 cm steak was removed (caudal end of the LT) from the grade site (12th to 13th rib), trimmed of all epimysium, subcutaneous and intermuscular fat, comminuted using a Robot Coupe Blixir BX3 (Robot Coupe USA Inc.; Ridgeland, MS, USA), and frozen at –80°C for subsequent fatty acid analyses. The remainder of the muscle was trimmed of all extraneous fat to the epimysium, weighed, vacuum packaged (Multivac AGW; Multivac Inc., Kansas City, MO) and aged for 13 d at 2°C. Following ageing period, two 2.5-cm steaks were then removed from the caudal end of the LT. Cooking losses and shear force values were determined on the first steak, and the second steak was stored at –20°C for 2 mo for further sensory analyses conducted by trained panelists. The remaining portion of the LT was trimmed of epimysium and comminuted using Robot Coupe Blixir BX3 (Robot Coupe USA Inc., Ridgeland, MS) for proximate analyses on raw samples.

The left *gluteus medius* (GM) muscle was also collected at 72 h post mortem and an 800-g subsample from the same location of lean muscle weighed, vacuum packaged and stored at 2°C. After 13 d of storage, the GM lean and perirenal fat from each steer were ground separately (Butcher Boy Meat Grinder, Butcher Boy Machines, Selmer, TN), initially with a 6-mm grind plate followed by a second grind using a 4 mm grind plate to achieve an 80/20 lean to fat grind. The grind was subsampled, further comminuted and frozen at –80°C for subsequent fatty acid analyses. The remaining grind was used to form 140 g patties (11.4 cm diameter × 0.64 cm thick) using a single patty hamburger press (Cabelas, Sydney, NE). Hamburgers were frozen at –20°C for 2 mo until sensory analyses were performed by trained panelists.

Fatty acid analyses

Fatty acid composition from LT and 80/20 lean to fat grind samples was analyzed by gas chromatography as described in Vahmani et al. (2017).

The peroxidizability index (PI) from LT and hamburgers, which shows the relative rate of lipid peroxidation reaction, was calculated from their fatty acid composition according to the following equation (Arakawa and Sagai, 1986): PI = (% monoenoic × 0.025) + (% dienoic × 1) + (% trienoic × 2) + (% tetraenoic × 4) + (% pentaenoic × 6).

Meat quality analyses

Steak weights were recorded prior to cooking. A 10-cm spear point Type T thermocouple probe (Wika Instruments, Edmonton, AB, Canada) was inserted into the center of the steak, and connected to a Hewlett-Packard HP34970A Data Logger (Hewlett-Packard Co., Boise ID) to monitor the internal temperature while cooking steaks. Steaks were grilled to an internal temperature of 35.5°C, flipped, and were removed when they reached 71°C, on a Garland grill (Model ED30B, Condon Barr Food Equipment Ltd., Edmonton, AB) that was preheated to 210°C. Upon removal from the grill, steaks were sealed in polyethylene bags and immersed in an ice-water bath to avoid further cooking. Cooked steaks were then chilled at 2°C for 24 h. After the chilling, steak weights were recorded again and six 1.9 cm diameter cores were cut out parallel to the fiber grain. Core peak shear force was determined perpendicular to the fiber grain (TA-XT Plus Texture Analyzer equipped with a Warner-Bratzler shear head at a crosshead speed of 200 mm × min^{-1} and a 30 kg load cell using Texture Exponent 32 Software; Texture Technologies Corp., Hamilton, MA). Shear force (kg) at 13 d for all steaks was calculated as the average of all six cores. Final and precooked steak weights were used to calculate cooking losses (mg × g^{-1}) and cooking times (s × g^{-1}).

Moisture and fat content (method 2008.06; AOAC, 2008) were analyzed using rapid analysis systems (Smart Turbo Moisture Analyzer Model 907990, and Smart Trac Fat Analyzer Model 907955; CEM Corporation, Matthews, NC). Protein content (method 2011.04; AOAC, 2011), was analyzed using an Elementar Rapid N Cube (Elementar Analysensysteme GmbH, Hanau, Germany).

Sensory analyses

After thawing steaks at 4°C for 24 h, they were cooked in the same manner described above. Following removal from the grill, steaks were cooled for 3 min, and then each steak was subsampled by cutting 1.3 × 1.3 × 1.3 cm cubes attempting to avoid areas with high levels of fat or connective tissue. The temperature of the steak cubes was equilibrated prior to sensory analysis by placing samples in covered glass containers in a circulating water bath (68°C). Samples were presented to a 9-member trained expert meat evaluation panel in a balanced design with sample assignment completed by Compusense 5 Software, version 4.6 (Compusense Inc., Guelph, ON). Attribute ratings were collected electronically. The panelists rated the following attributes from the steak samples: initial and overall ten-

derness, initial and sustained juiciness, beef flavor and off-flavor intensity, amount of connective tissue, and residual mouth coating. The scores were assigned using nine-point descriptive scales: 9 = extremely tender, extremely juicy, extremely intense beef flavor, extremely bland off-flavor, no connective tissue, and no residual mouth coating; 1 = extremely tough, extremely dry, extremely bland beef flavor, extremely intense off-flavor, extremely abundant connective tissue, and extremely abundant residual mouth coating. Panelists evaluated the off-flavor (metallic, off-sour, livery, grainy, bloody/serumy, unidentified, other, and none) and texture (typical beef, mushy, mealy, spongy, rubbery, and crumbly) descriptors. As well, panelists were asked to identify the 'other' off-flavor by free choice profiling.

Paid panelists were externally recruited, screened and trained (American Meat Science Association, 2016) to exclusively evaluate meat samples and have served on the panel as trained experts for an average of 6 yr. Evaluation and monitoring of panelists' performance were also performed according to the guidelines of American Meat Science Association (2016) for descriptive testing. The number of samples evaluated per session was 6. In total, 4 sessions were conducted per day: 2 in the morning and 2 in the afternoon, with a 20 min break between sessions. All panel evaluations were conducted in well-ventilated, partitioned booths, illuminated by 180 lux green lighting. Distilled water and unsalted soda crackers were provided to cleanse the palate of residual flavor notes between samples (Larmond, 1977).

Statistical analyses

The effect of diet on meat quality and sensory characteristics was determined by one-way analysis of variance (ANOVA), with pen as experimental unit, using the MIXED procedure in the Statistical Analysis Software system (Version 9.2, SAS Inst. Inc., Cary, NC). Sensory data analysis also included trained panelist as a random effect. Chi-square analysis was used to analyze the frequency of off-flavor and texture descriptors. Least square means differences were identified as significant at $P < 0.05$ using the PDIFF option in SAS.

Partial least squares discriminant analysis (PLS-DA) based on fatty acids was applied to classify steaks into presence or absence of fishy off-flavor (Fishy and Non-Fishy, respectively). This model attempts to correlate fatty acid variations (X) with defined classes (Y), while maximizing the covariance between the two types of variables for group differences and ignoring in-class variance. In this type of approach, Y (Off-flavor) is a dummy matrix with arbitrary numbers assigned to the different classes (Non-Fishy = 1; Fishy = 2; Naes et al., 2002). Cross-validation (leave one-out) was conducted to validate calibration and to restrict the number of PLS terms incorporated in the regression, to prevent overfitting. Correlation loadings from the PLS-DA were calculated to identify the fatty acids significantly ($P < 0.05$) correlated with the presence of fishy off-flavor. PLS-DA was conducted using The Unscrambler software (version 10.2, Camo, Trondheim, Norway).

Results and Discussion

Effect of feeding flaxseed in TMR vs. non-TMR on steak eating quality

Quality and sensory characteristics of steaks from steers fed TMR compared to Non-TMR are shown in Table 1. Diet did not affect chemical composition or cooking losses ($P > 0.10$), but did affect shear force values of steaks. Steaks from steers fed Non-TMR required 1.01 kg more shear force ($P < 0.05$) than those fed TMR, probably due to a higher early post mortem pH (Marsh et al., 1987). As shown in Vahmani et al. (2017), steers fed Non-TMR had higher 45 min post-slaughter LT pH (6.79 vs. 6.69, $P = 0.01$) and tended to have higher 3 d post-slaughter LT pH (5.57 vs. 5.51, $P = 0.07$) than those fed TMR. A lower dry matter intake at the end of the trial in the Non-TMR steers (10.6 vs. 11.4 kg DM.d^{-1}, $P < 0.05$, Vahmani et al., 2017) could result in lower LT glycogen reserves prior to slaughter. Hence, lactic acid production could have been reduced leading to slightly higher meat pH.

A difference in shear force of 1 kg can be typically detected by panelists (Aalhus et al., 2004), and in this experiment, trained panelists found tendencies for lower (less tender) initial and overall tenderness scores ($P = 0.09$ and 0.07, respectively) for Non-TMR compared to TMR steaks. No significant differences were found by panelists for juiciness, amount of connective tissue or residual mouth coating ($P > 0.10$), which agree with the lack of significant differences in quality grades between steaks from TMR and non-TMR steers (Vahmani et al., 2017). However, Non-TMR steaks had significantly lower beef flavor intensity (–0.62 panel units, $P < 0.01$) and greater off-flavor intensity (lower off-flavor values, –1.30 panel units, $P < 0.01$) than those from TMR steers. This agrees with previous studies (Hernández-Calva et al., 2011; LaBrune et al., 2008), who found an increase of off-flavor intensity in beef when feeding flaxseed. Conversely, McNiven et al. (2011) reported no effect

Table 1. Quality and sensory characteristics of steaks and hamburgers from steers fed extruded flaxseed and ground hay either together as a total mixed ration (TMR) or sequentially (Non-TMR)

	Non-TMR		TMR		
Parameter	Mean	SEM[1]	Mean	SEM	*P*-value
Steak					
Quality characteristics					
Cooking loss, mg × g^{-1}	216.71	7.44	207.18	7.44	0.37
Cooking time, s × g^{-1}	5.29	0.26	4.96	0.26	0.36
Shear force, kg	6.11[a]	0.28	5.10[b]	0.28	< 0.05
Moisture, %	70.24	0.43	70.25	0.43	0.98
Fat, %	7.40	0.57	7.49	0.57	0.91
Protein, %	21.35	0.16	21.25	0.16	0.65
Sensory attributes[2]					
Initial tenderness	6.17	0.24	6.68	0.24	0.09
Initial juiciness	5.75	0.17	5.66	0.17	0.63
Beef flavor intensity	4.33[b]	0.30	4.95[a]	0.30	< 0.01
Off-flavor intensity	6.20[b]	0.34	7.50[a]	0.34	< 0.01
Amount of connective tissue	8.05	0.28	8.17	0.28	0.41
Overall tenderness	6.54	0.20	7.01	0.20	0.07
Sustained juiciness	5.74	0.22	5.70	0.22	0.65
Residual mouth coating	7.30	0.21	7.59	0.21	0.13
Hamburger					
Sensory attributes[2]					
Initial tenderness	7.39	0.20	7.40	0.20	0.84
Initial juiciness	5.54	0.18	5.63	0.18	0.60
Beef flavor intensity	5.01	0.23	5.02	0.23	0.98
Off-flavor intensity	7.08	0.30	7.35	0.30	0.27
Overall tenderness	7.53	0.26	7.59	0.26	0.19
Sustained juiciness	5.62	0.19	5.79	0.19	0.29
Residual mouth coating	6.40	0.17	6.42	0.17	0.90

[a,b]Within row, means without a common superscript differ ($P < 0.05$).

[1]SEM: standard error of least square means.

[2]Nine point descriptive scale for initial and overall tenderness (9 = extremely tender; 1 = extremely tough), initial and sustained juiciness (9 = extremely juicy; 1 = extremely dry), beef flavor intensity (9 = extremely intense; 1 = extremely bland), off-flavor intensity (9 = no off-flavor; 1 = extremely intense off-flavor), amount of connective tissue (9 = none detected; 1 = extremely abundant) and residual mouth coating (9 = none residual mouth coating; 1 = extremely abundant residual mouth coating).

of feeding flaxseed on freshly cooked beef off-flavor. Differences in production systems, animal management systems, and diets have produced different findings in various studies in terms of the relationship between fatty acid composition and flavor (Calkins and Hodgen, 2007).

When the panelists evaluated the off-flavor descriptors (Table 2), a significantly ($P < 0.01$) higher percentage of panelists indicated some off-sour in the TMR steaks compared to the Non-TMR. Different substrates in meat, due to differences in pH and fatty acid profile between the TMR and Non-TMR steaks (Vahmani et al., 2017), might have caused different rates of proliferation and/or different strains of lactic acid bacteria in steaks from both treatments, which are known for the development of a sour off-flavor (Egan, 1983). Despite those off-sour notes, absence of off-flavor (None) was most frequently reported by the panelists for the TMR steaks (43.1%, $P <$ 0.0001), whereas a significantly high proportion of panelists indicated some off-flavor in the Non-TMR (48.3%, $P < 0.0001$). The off-flavor found in the Non-TMR was different from the classic off-flavors described in the beef lexicon for intact muscle by Adhikari et al. (2011) and was identified as 'other' accordingly. When the panelists were asked to identify that 'other' off-flavor from the Non-TMR steaks, 69% of the panelists described it as fishy, and 19% as oily. In steers fed fish oils, fishy off-flavor has been shown to occur more with higher levels of long chain PUFA, as these fatty acids are more likely to become oxidized (Scollan et al., 2006). Similarly, Lee et al. (2009) indicated that the fishy off-flavor found in cattle fed red clover silage may be related to the elevated levels of long chain fatty acids and overall n-3 PUFA observed in the meat. In this study, the concentrations of long-chain n-3 PUFA in LT such as 20:5n-3, 22:5n-3 and

Table 2. Frequency (percentage of respondents) for off-flavor and texture descriptors of steaks from steers fed extruded flaxseed and ground hay either together as a total mixed ration (TMR) or sequentially (Non-TMR)

Descriptors	Non-TMR	TMR	*P*-value[1]
Off-flavor descriptors			
Metallic	0	0.43	–
Off-sour	7.26	15.95	< 0.01
Livery	0.43	2.16	0.10
Grainy	2.56	0.43	0.06
Bloody/Serumy	2.14	3.02	0.56
Unidentified	18.38	21.55	0.47
None	20.94	43.10	< 0.0001
Other	48.29	13.36	< 0.0001
Coconut Oil	0.43	0.86	
Fishy	33.33	5.60	
Flaxy	3.42	0.43	
Oily	9.40	4.31	
Stale	1.71	1.72	
Sweet	0.00	0.43	
Texture descriptors			
Typical	57.26	44.02	< 0.05
Mushy	20.51	34.19	< 0.01
Mealy	3.85	6.41	0.22
Spongy	6.41	5.13	0.56
Rubbery	6.41	4.27	0.32
Crumbly	5.56	5.98	0.85

[1]Chi-square frequencies were significant at $P < 0.05$.

22:6n-3 were not different between treatments ($P > 0.10$; Vahmani et al., 2017). Nevertheless, the Non-TMR steaks had significantly higher proportion of 18:3n-3 and many of its BHI products ($P < 0.01$), some of which are known to be relatively unstable (ex. conjugated linoleic acid; Zhang and Chen, 1997). Hence, a detailed study of the relationship between individual fatty acids and the fishy off-flavor found in the Non-TMR steaks was undertaken.

To determine the fatty acids associated with the fishy off-flavor observed in the Non-TMR steaks, a PLS-DA was performed based on individual fatty acid proportions. One steak was considered as having a fishy off-flavor when 3 or more panelists (out of 9 panelists) detected that off-flavor in the steak. Figure 1a shows the score plot from the PLS-DA performed based on the fatty acids to classify steaks according to presence (Fishy) or absence (Non-Fishy) of fishy off-flavor. With the entire sample set represented on an XY plane according to the scores for PC 1 and PC 2, the regression model developed including only 2 PLS terms identified 2 clusters: the first grouped most of the Fishy samples and the second cluster contained the Non-Fishy samples. Hence, a clear separation between Fishy and Non-Fishy was observed according to the fatty acid composition, with only 1 sample considered as Fishy by the panelists being classified into the Non-Fishy cluster. It is worth mentioning that this 'misclassified' sample was the only steak from the steers fed TMR in which panelists detected the fishy off-flavor (Fig. 1b), because the rest of TMR steaks were considered as Non-Fishy and all the Non-TMR steaks as Fishy. This suggests that, despite having a fishy off-flavor, the overall fatty acid profile of that sample was similar to the rest of the TMR steaks based on the score plot, and was classified into the same cluster (TMR) accordingly. Similar results were observed when the calibration model was cross-validated, where 2 clusters (Fishy and Non-Fishy) were clearly identified based on the fatty acid profile, with only 1 sample misclassified (graph not shown).

The fatty acids significantly correlated with the presence of fishy off-flavor are circled (⦿) in Fig. 2, where the correlations between each fatty acid proportion and the 2 first dimensions (PC1 and PC 2) were depicted as correlation loadings, with concentric circles of radii representing 50% and 100% of the explained variance (inner and outer ellipse, respectively). As observed, the correlation loading plot revealed significant high loadings (outer ellipse) for most of the *trans* monounsaturated fatty acids (*t*-MUFA), *cis*(*c*)-MUFA, CLnA, conjugated linoleic acid (CLA) isomers and atypical dienes (AD, i.e., 18:2 isomers that are not linoleic or CLA). Specifically, *t*-MUFA (*t*11–18:1; *t*9–16:1), CLA (*c*9,*t*11–18:2; *t*11,*c*13–18:2), CLnA (*c*9,*t*11,*t*15–18:3; *c*9,*t*11,*c*15–18:3) and AD (*t*11,*t*15–18:2; *t*11,*c*15–18:2; *t*9,*c*12–18:2; *t*9,*t*12–18:2) were positioned on the right quadrant near the 'Fishy off-flavor', hence being positively correlated with the presence of this off-flavor. In contrast, *c*-MUFA with significant high loadings such as *c*12–18:1, *c*14–18:1 and *c*16–18:1 were negatively correlated with the fishy off-flavor (left quadrant of the correlation loading plot). These results are in agreement with Nuernberg et al. (2005), who found significantly more fishy off-flavor and lower overall flavor desirability in meat from grass-finished cattle with increased *t*-18:1 isomers and, notably, the CLA isomer *c*9,*t*11–18:2. In this study, *c*9,*t*11–18:2 did not have a high correlation loading (inner ellipse) but *t*11,*c*13–18:2 had high correlation loading (outer ellipse), and both had enough structured variation to be positively correlated ($P < 0.05$) to the fishy off-flavor. Indeed the Non-TMR steaks had significantly higher contents of these CLA isomers when compared to TMR (Vahmani et al., 2017). All the individual AD fatty acids appeared circled in the correlation loading plot, hence being significant for the steak clustering based on the presence or absence of fishy off-flavor. Indeed, feeding Non-TMR significantly increased ($P < 0.01$)

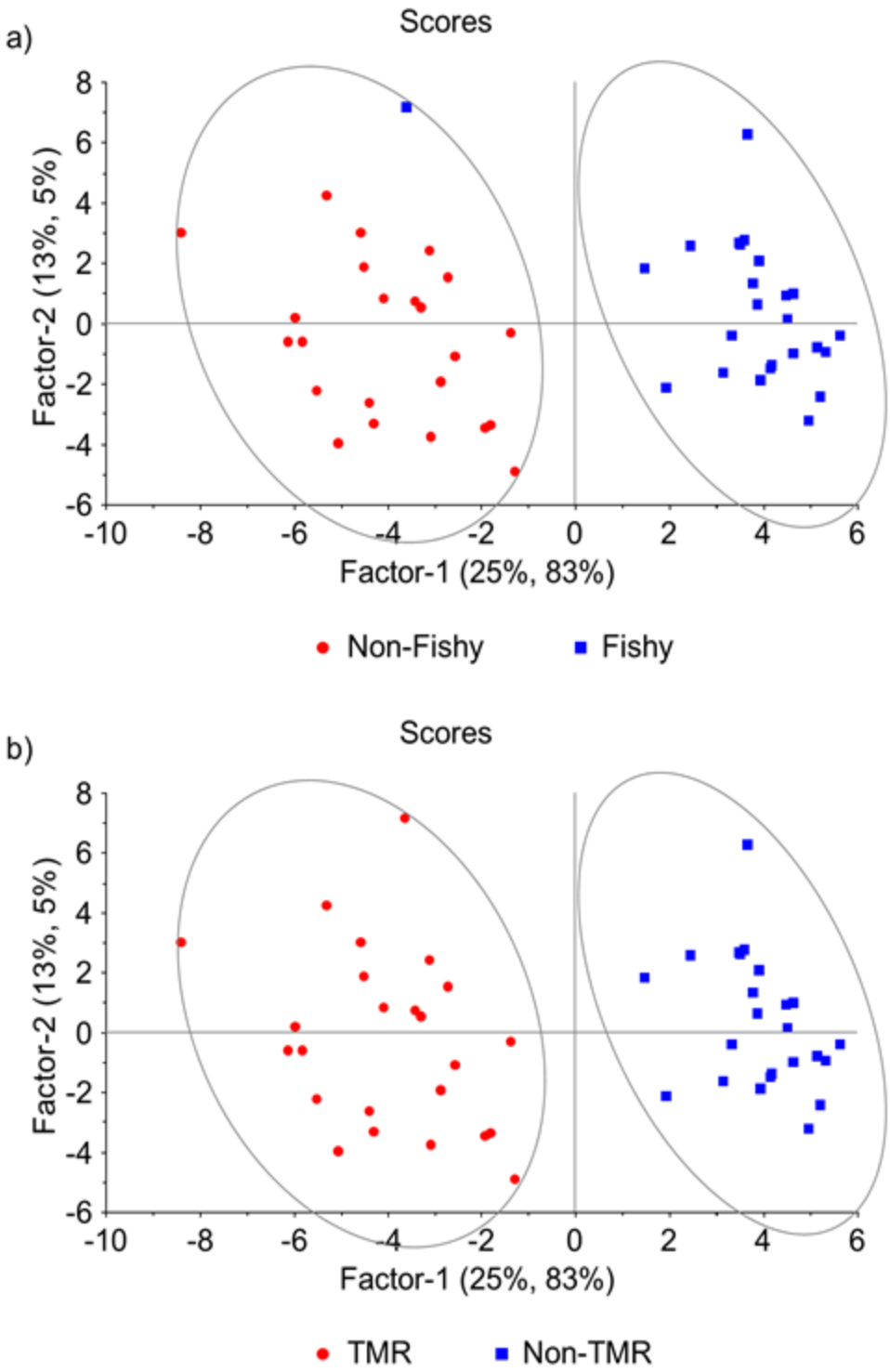

Figure 1. a) Score plot based on principal component (PC) 1 and PC 2 from the partial least squares discriminant analysis (PLS-DA) using the fatty acid profile (%) to classify steaks based on the presence or absence of fishy off-flavor. b) TMR (total mixed ration) and Non-TMR selected as sample grouping in the score plot from the PLS-DA.

proportions of total AD in LT, mainly of *t*11,*c*15–18:2 (Vahmani et al., 2017). Data on the oxidative stability of AD fatty acids is, however, lacking.

Results from individual fatty acids were similar to those obtained when a PLS-DA was performed with the proportions of fatty acid groups to classify steaks based on the presence (Fishy) or absence (Non-Fishy) of fishy off-flavor. As observed in the correlation loading plot (Fig. 3), total *t*-MUFA presented a high correlation loading (outer ellipse) and were significantly correlated with

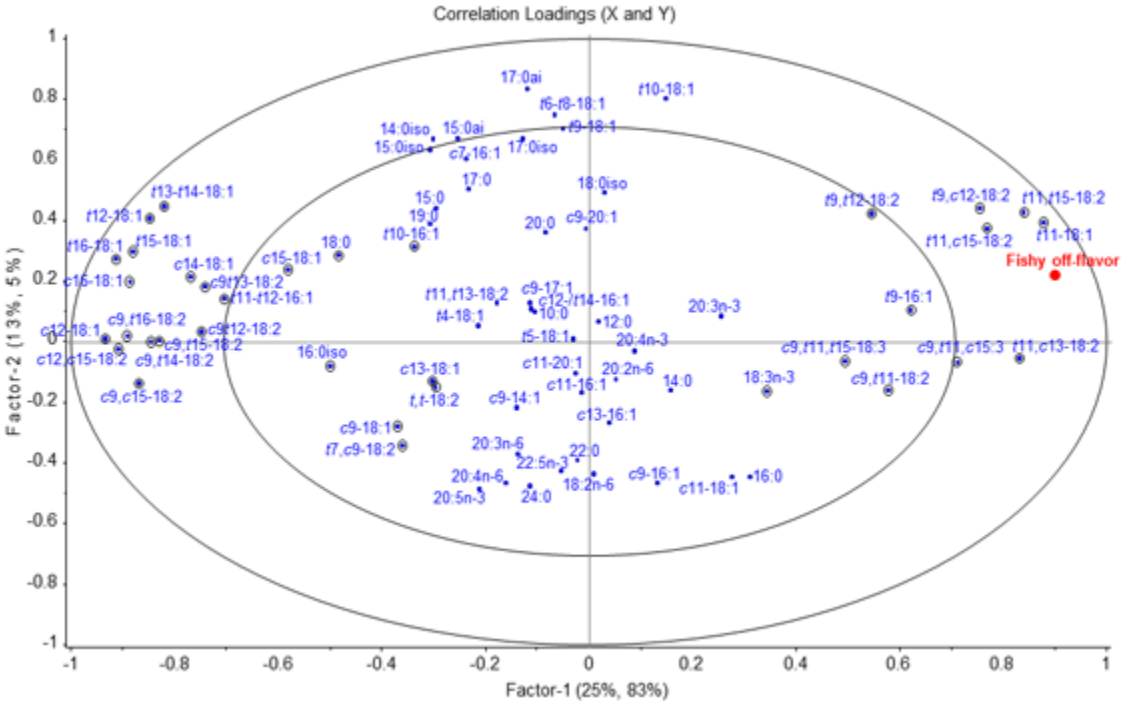

Figure 2. Correlation loading plot from the partial least squares discriminant analysis showing the individual fatty acids positively and negatively correlated with the presence of fishy off-flavor. Circled (⦿) individual fatty acids were significant at $P < 0.05$.

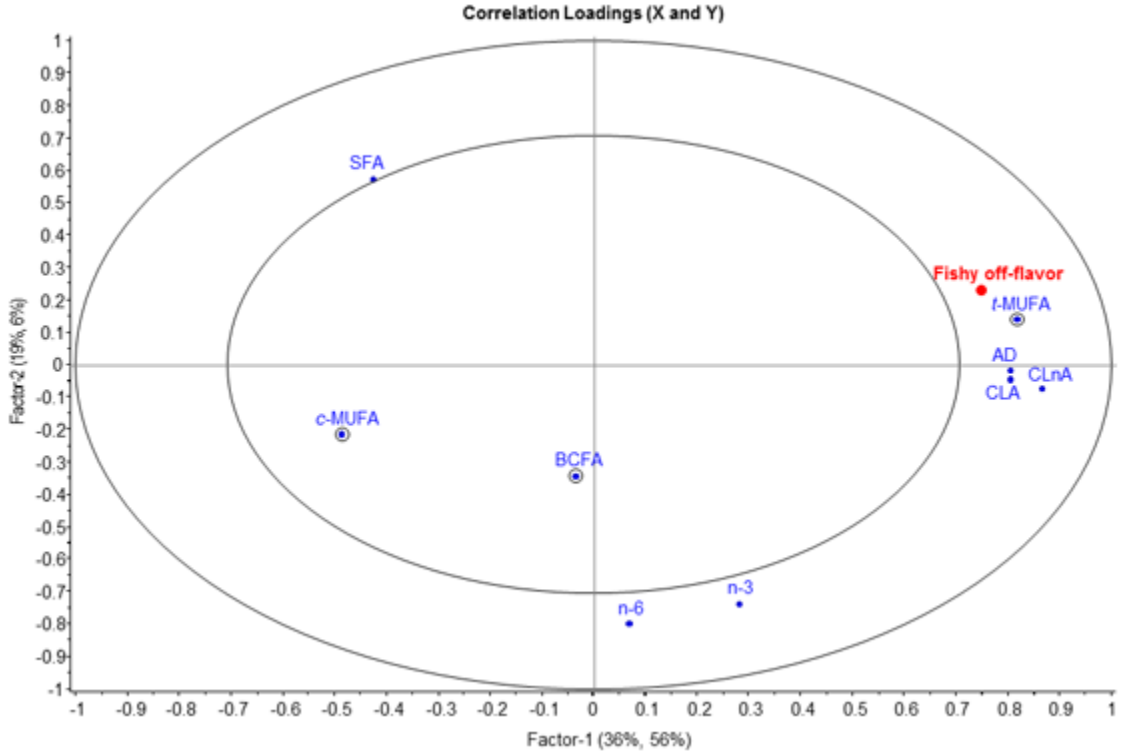

Figure 3. Correlation loading plot from the partial least squares discriminant analysis showing the fatty acid groups positively and negatively correlated with the presence of fishy off-flavor. Circled (⦿) fatty acid groups were significant at $P < 0.05$. AD: atypical dienes; BCFA: branched chain fatty acids; CLA: conjugated linoleic acid; CLnA: conjugated linolenic acid; *c*-MUFA: *cis*-mounsaturated fatty acids; *t*-MUFA: *trans*-mounsaturated fatty acids; SFA: saturated fatty acids.

the presence of fishy off-flavor. They were positioned on the right quadrant near the 'Fishy off-flavor', hence being positively correlated with this off-flavor. Albeit significant, *c*-MUFA presented a lower correlation loading (inner ellipse) and were negatively correlated (left quadrant) to the fishy off-flavor. Total CLA, CLnA and AD were not significant, although presented a high correlation loading (outer ellipse) and were positively correlated with the fishy off-flavor. The lack of significance for total AD likely relates to higher proportions of AD in Non-TMR steaks except for several Δ9-desaturation products of *t*18:1 isomers found at higher concentrations in TMR steaks. In contrast, total branched chain fatty acids (BCFA) were significant, despite only 16:0iso was significantly correlated with the fishy off-flavor (Fig. 2). Nevertheless, the correlation loading for this group of fatty acids was low (inner ellipse). In cooked mutton, undesirable flavors have been in part attributed to branched-chain fatty acids with 8 to 10 carbon atoms (Wong et al., 1975). However, no studies establishing clear relationships between long carbon chain (> 10) BCFA and off-flavors in beef were found in the literature.

From these results and results of others, we could conclude that the *t*-MUFA are associated with a fishy off-flavor in beef. Within this group, the most abundant fatty acid (*t*11–18:1) presented the highest positive correlation loading with regard to the fishy off-flavor (Fig. 2). Similarly, when the regression coefficients from the PLS-DA were plotted (graph not shown), the vaccenic acid presented the highest coefficient of regression. Indeed, feeding Non-TMR significantly increased the proportions of total *t*-MUFA and vaccenic acid in the steaks compared to TMR (11.36 vs. 9.73%, 7.43% vs. 4.97%, respectively; $P < 0.0001$; Vahmani et al., 2017). Reasons for those differences between TMR and non-TMR are unclear, but could in part relate to differences in rumen microbial populations, which could in turn result in different biohydrogenation pathways (Jenkins et al., 2008). These results agree with Camfield et al. (1997), who found that the vaccenic acid was involved in the development of off-flavors in beef. In the current study, every Non-TMR steak presented greater proportions of vaccenic acid (≥ 6.12% of total fatty acids) when compared to the TMR. There was only 1 TMR steak with a high vaccenic acid content (6.51% vaccenic of total fatty acids). Not unexpectedly, that steak was the one considered as Fishy by the panelists despite being a TMR steak and, therefore, grouped together with all the TMR in the PLS-DA based on the fatty acid profile (Fig. 1). Hence, the fishy off-flavor might be associated with the unusually high level of vaccenic acid presented in that TMR steak. From these results, we could conclude that beef samples with levels of vaccenic acid ≥ 6.12% of total fatty acids resulting from feeding flaxseed products, while of potentially greater health benefit, may pose a risk of fishy off-flavor. Nevertheless, despite the association of proportion of vaccenic acid with the fishy off-flavor, this fatty acid is likely not directly causing the off-flavor as *t*18:1 are known to be relatively stable (Moser, 2009). The mechanism involved in the development of the fishy off-flavor may, therefore, be related to conjugated fatty acids and AD positively correlated with vaccenic acid content (CLnA, some CLA isomers, *t*11,*c*15–18:2 and *t*11,*t*15–18:2). The lack of a direct effect of vaccenic acid on oxidative stability was supported by the finding that not all the *t*-MUFA isomers were significantly correlated with the fishy off-flavor and some, while being significant, were negatively correlated with that off-flavor (left quadrant of the correlation loading plot, Fig. 2). Nevertheless, due to the great influence of vaccenic acid, 78% of the steaks with a proportion of total *t*-MUFA higher than 10% of total FA were detected as Fishy by the trained panelists.

Regarding texture descriptors (Table 2), both Non-TMR and TMR steaks had a high proportion of responses (about 50%) indicating that their predominant texture was that from typical beef. Nevertheless, the respondents indicating that texture descriptor was significantly higher ($P < 0.05$) in the Non-TMR. Steaks from both Non-TMR and TMR were also described as mushy, especially the TMR whose mushy frequency was significantly higher ($P < 0.01$) than for the Non-TMR. Low-fat meat products or meat with not enough connective tissue have been found as mushy (Barbut, 2011; Keeton, 1994). Additionally, BCFA seem to have a softening effect on lamb fat (Johnson et al., 1988). In this study, no statistically significant differences were found in the intramuscular fat content or amount of connective tissue detected by the panelists between the Non-TMR and TMR steaks (Table 1). Nevertheless, the total content of BCFA tended to increase in the TMR steaks ($P = 0.08$; Vahmani et al., 2017) when compared to the Non-TMR. Hence, the higher proportion of these fatty acids might have contributed to the greater mushy perception in the TMR steaks.

Effect of feeding flaxseed in TMR vs. Non-TMR on hamburger eating quality

When the eating quality of hamburgers manufactured with GM and perirenal fat (80/20) was evaluated, no statistical differences were found in any of the sensory attributes evaluated by the panelists between hamburgers from steers fed TMR or Non-TMR (Table 1).

Off-flavor and texture descriptors of hamburgers from steers fed TMR compared to Non-TMR are shown in Table 3. Absence of off-flavor was frequently reported by the panelists for both the TMR and Non-TMR hamburgers (34.3 and 42.2%, respectively). Nevertheless, a similar high proportion of panelists also observed some off-sour flavor in hamburgers from both treatments (35.3 and 31.9%, respectively). The off-sour flavor could be due to the storage period of both GM and perirenal fat, since both tissues were vacuum packaged and stored at 2°C for 13 d before hamburgers were manufactured. It is well known that under anaerobic conditions, lactic acid bacteria–known for the development of a sour "off" flavor– may become the dominant component of the microbial flora of meats with normal pH (Egan, 1983). Additionally, when compared to TMR hamburgers, a significantly higher number of panelists detected 'other' off-flavor in the Non-TMR (20.1 vs. 11.3%, $P < 0.05$). When the panelists were asked to identify that 'other' off-flavor by free choice profiling, almost half of the panelists (39%) identified that off-flavor as fishy. Indeed, the Non-TMR hamburgers had significantly higher proportions of vaccenic acid and total *t*-MUFA compared to the TMR ($P < 0.0001$, Vahmani et al., 2017); the vaccenic acid proportion averaged 9.5% of total fatty acids for Non-TMR hamburgers. Despite the significantly lower 'other' off-flavor for the TMR hamburgers, some fishy off-flavor was also observed (4.4% of the panelists). This could be because some TMR hamburgers showed high levels of vaccenic acid (TMR vaccenic acid average of 6.8% of total fatty acids, Vahmani et al., 2017), as a consequence of high levels of this fatty acid in perirenal fat. In addition, 5.4% of the panelists detected some stale/cardboard off-flavor in the Non-TMR hamburgers, and also in the TMR but to a lesser extent (2.9% of the panelists). Therefore, although fishy was reported as the predominant off-flavor in the Non-TMR hamburgers, the presence of that off-flavor was not as definitive as in the Non-TMR steaks (7.8% vs. 33.3% of the panelists). Hence, the fishy off-flavor might have been masked by other off-flavors presented in the Non-TMR hamburgers such as stale/cardboard. The presence of other off-flavors in hamburgers compared to the steaks could be expected, since precooked ground meat products are very susceptible to lipid oxidation, which can lead to undesirable flavor and odor changes during storage (Kanner, 1994). This may be due to the high fat content (20 to 30%) and the processes which precooked ground meats are subjected (Kanner, 1994). Secondary compounds produced from the oxidation of PUFA contribute to warmed-over-flavors (stale, wet cardboard, painty, grassy, rancid) observed in cooked, stored beef (Campo et al., 2006); in particular, cardboard off-flavor is found early in the lipid oxidation process (Angelo et al., 1990).

Table 3. Frequency (percentage of respondents) for off-flavor and texture descriptors of hamburgers from steers fed extruded flaxseed and ground hay either together as a total mixed ration (TMR) or sequentially (Non-TMR)

Descriptors	Non-TMR	TMR	*P*-value[1]
Off-flavor descriptors			
Metallic	0.49	0	–
Off-sour	35.29	31.86	0.55
Livery	0.49	0	–
Grainy	0	0.98	–
Bloody/Serumy	0	1.47	–
Unidentified	9.31	12.25	0.37
None	34.31	42.16	0.20
Other	20.10	11.27	< 0.05
Barny	0.49	0.49	
Citrusy	0.49	0.49	
Fishy	7.84	4.41	
Flaxy	0.98	0.00	
Fruity	1.47	0.00	
Musky	0.49	0.00	
Oily	1.47	2.45	
Salty	0.98	0.49	
Stale/Cardboard	5.39	2.94	
Sweet	0.49	0.00	
Texture descriptors			
Typical	87.25	81.37	0.52
Mushy	4.41	8.33	0.12
Mealy	3.92	5.39	0.49
Spongy	0	0	–
Rubbery	0	0	–
Crumbly	4.41	4.90	0.82

[1]Chi-square frequencies were significant at $P < 0.05$.

Since the number of double bonds in a fatty acid affects the susceptibility to oxidation, the different frequency of 'other' off-flavor found in the hamburgers from this study could be due to differences in the content of long chain PUFA and amount of oxidation. In this regard, Non-TMR hamburgers had significantly higher concentrations of relatively unstable PUFA, such as CLA, CLnA, and AD than the TMR ($P < 0.001$, Vahmani et al., 2017). Likewise, the peroxidizability index was higher in the Non-TMR than in the TMR hamburgers (13.59 vs. 11.48%, $P < 0.0001$), indicating a higher relative rate of lipid peroxidation in the non-TMR.

Overall, the results from this study agree with Turner et al. (2015), who observed an increase in off-flavor intensity and oxidation in hamburgers manufactured with perirenal fat from cattle fed flaxseed. Additionally, Jiang et al. (2011) observed that increasing levels of long-chain

n-3 PUFA in ground beef patties had negative effects on sensory quality, especially on off-flavor. Regarding texture descriptors, no differences were found between the Non-TMR and TMR hamburgers, since most of the panelists indicated that both had an expected typical texture of hamburgers (87.2 and 81.4%, respectively).

Conclusions

Although feeding flaxseed in a Non-TMR compared to TMR enhanced the deposition of α-linolenic acid and its BHI in beef, it resulted in tougher steaks with lower beef flavor and greater off-flavor intensity to the extent where these might be detectable by consumers. Beef samples with a threshold level of vaccenic acid over 6.12% of total fatty acids resulting from feeding flaxseed products presented a fishy off-flavor that may compromise eating quality, although acceptability would need to be tested with a consumer panel. Further research is required to study the effects of greater levels of antioxidant protection (i.e., from feeding vitamin E) than provided in the present trial and/or protective packaging, and the effect of ageing on changes in antioxidant capacity, oxidative stability and shelf life in beef with enhanced fatty acid profiles.

Literature Cited

Aalhus, J. L., L. E. Jeremiah, M. E. R. Dugan, I. L. Larsen, and L. L. Gibson. 2004. Establishment of consumer thresholds for beef quality attributes. Can. J. Anim. Sci. 84(4): 631–638. doi:10.4141/A03-106

Adhikari, K., E. Chambers, IV, R. Miller, L. Vázquez-Araújo, N. Bhumiratana, and C. Philip. 2011. Development of a lexicon for beef flavor in intact muscle. J. Sens. Stud. 26(6):413–420. doi:10.1111/j.1745-459X.2011.00356.x

American Meat Science Association. 2016. Research guidelines for cookery, sensory evaluation, and instrumental tenderness measurements of meat. Am. Meat Sci. Assoc., Chicago, IL.

Angelo, A. J. S., K. L. Crippen, H. P. Dupuy, and C. James. 1990. Chemical and sensory studies of antioxidant-treated beef. J. Food Sci. 55(6):1501–1505. doi:10.1111/j.1365-2621.1990.tb03554.x

AOAC. 2008. Official method for moisture and fat in meats microwave and nuclearmagnetic resonance analysis [2008.06]. Official methods of analysis. AOAC International, Gaithersburg, MD.

AOAC. 2011. Official method for protein in raw and processed meats [2011.04]. Official methods of analysis. AOAC International, Gaithersburg, MD.

Arakawa, K., and M. Sagai. 1986. Species differences in lipid peroxide levels in lung tissue and investigation of their determining factors. Lipids 21(12):769–775. doi:10.1007/BF02535410

Barbut, S. 2011. Reducing fats in processed meat products In: J. P. Kerry and J. F. Kerry, editors, Processed meats improving safety, nutrition and quality. Woodhead Publishing Limited, Cambridge, UK. p. 346–366.

Benjamin, S., and F. Spener. 2009. Conjugated linoleic acids as functional food: An insight into their health benefits. Nutr. Metab. 6(1):36. doi:10.1186/1743-7075-6-36

Calkins C. R., and Hodgen J. M. 2007. A fresh look at meat flavor. J. Meat Sci. 77(1):63–80. doi:10.1016/j.meatsci.2007.04.016

Camfield, P. K., A. H. Brown, P. K. Lewis, L. Y. Rakes, and Z. B. Johnson. 1997. Effects of frame size and time-on-feed on carcass characteristics, sensory attributes, and fatty acid profiles of steers. J. Anim. Sci. 75(7):1837–1844. doi:10.2527/1997.7571837x

Campo, M. M., G. R. Nute, S. I. Hughes, M. Enser, J. D. Wood, and R. I. Richardson. 2006. Flavour perception of oxidation in beef. Meat Sci. 72(2):303–311. doi:10.1016/j.meatsci.2005.07.015

CCAC. 2009. Canadian Council on Animal Care guidelines on the care and use of farm animals in research, teaching and testing. Ottawa, ON. Http://www.ccac.ca/Documents/Standards/Guidelines/Farm_Animals.pdf (accessed 24 August 2017).

Egan, A. F. 1983. Lactic acid bacteria of meat and meat products. Antonie van Leeuwenhoek 49(3):327–336. doi:10.1007/BF00399507

Elmore, J. S., M. M. Campo, M. Enser, and D. S. Mottram. 2002. Effect of lipid composition on meat-like model systems containing cysteine, ribose, and polyunsaturated fatty acids. J. Agric. Food Chem. 50(5):1126–1132. doi:10.1021/jf0108718

Field, C. J., H. H. Blewett, S. Proctor, and D. Vine. 2009. Human health benefits of vaccenic acid. Appl. Physiol. Nutr. Metab. 34(5):979–991. doi:10.1139/H09-079

Hernández-Calva, L. M., M. He, M. Juárez, J. L. Aalhus, M. E. R. Dugan, and T. A. McAllister. 2011. Effect of flaxseed and forage type on carcass and meat quality of finishing cull cows. Can. J. Anim. Sci. 91:613–622. doi:10.4141/cjas2011-030

Jenkins, T. C., R. J. Wallace, P. J. Moate, and E. E. Mosley. 2008. Board-invited review: Recent advances in biohydrogenation of unsaturated fatty acids within the rumen microbial ecosystem1. J. Anim. Sci. 86(2):397–412. doi:10.2527/jas.2007-0588

Jiang, T., J. R. Busboom, M. L. Nelson, and R. Mengarelli. 2011. Omega-3 fatty acids affected human perception of ground beef negatively. Meat Sci. 89(4):390–399. doi:10.1016/j.meatsci.2011.04.026

Johnson, C. B., R. W. Purchas, and E. J. Birch. 1988. Fatty acid composition of fats of differing melting points extracted from ram subcutaneous tissue. Lipids 23(11):1049–1052. doi:10.1007/BF02535650

Kallas, Z., C. E. Realini, and J. M. Gil. 2014. Health information impact on the relative importance of beef attributes including its enrichment with polyunsaturated fatty acids (omega-3 and conjugated linoleic acid). Meat Sci. 97(4):497–503. doi:10.1016/j.meatsci.2014.03.015

Kanner, J. 1994. Oxidative processes in meat and meat products: Quality implications. Meat Sci. 36:169–189. doi:10.1016/0309-1740(94)90040-X

Keeton, J. T. 1994. Low-fat meat products—technological problems with processing. Meat Sci. 36:261–276. doi:10.1016/0309-1740(94)90045-0

Kim, J. H., Y. Kim, Y. J. Kim, and Y. Park. 2016. Conjugated linoleic acid: Potential health benefits as a functional food ingredient. Annu. Rev. Food Sci. Technol. 7:221–244. doi:10.1146/annurev-food-041715-033028

Kronberg, S. L., G. Barceló-Coblijn, J. Shin, K. Lee, and E. J. Murphy. 2006. Bovine muscle n-3 fatty acid content is increased with flaxseed feeding. Lipids 41(11):1059–1068. doi:10.1007/s11745-006-5056-z

LaBrune, H. J., C. D. Reinhardt, M. E. Dikeman, and J. S. Drouillard. 2008. Effects of grain processing and dietary lipid source on performance, carcass characteristics, plasma fatty acids, and sensory properties of steaks from finishing cattle. J. Anim. Sci. 86:167–172. doi:10.2527/jas.2007-0011

Larmond, E. 1977. Laboratory methods for sensory evaluation of food. Agriculture Canada, Research Branch. Ottawa, Canada.

Lee, M. R. F., P. R. Evans, G. R. Nute, R. I. Richardson, and N. D. Scollan. 2009. A comparison between red clover silage and grass silage feeding on fatty acid composition, meat stability and sensory quality of the M. Longissimus muscle of dairy cull cows. Meat Sci. 81(4):738–744. doi:10.1016/j.meatsci.2008.11.016

Marsh, B. B., T. P. Ringkob, R. L. Russell, D. R. Swartz, and L. A. Pagel. 1987. Effects of early-postmortem glycolytic rate on beef tenderness. Meat Sci. 21(4):241–248. doi:10.1016/0309-1740(87)90061-1

McNiven, M. A., J. L. Duynisveld, T. Turner, and A. W. Mitchell. 2011. Ratio of n-6/n-3 in the diets of beef cattle: Effect on growth, fatty acid composition, and taste of beef. Anim. Feed Sci. Technol. 170(3–4):171–181. doi:10.1016/j.anifeedsci.2011.09.006

Miller, A. 2001. Beef flavor: A white paper. A paper prepared for the National Cattlemen's Beef Association, Centennial, CO.

Mottram, D. S. 1998. Flavour formation in meat and meat products: A review. Food Chem. 62(4):415–424. doi:10.1016/S0308-8146(98)00076-4

Moser, B. R. 2009. Comparative oxidative stability of fatty acid alkyl esters by accelerated methods. J. Am. Oil Chem. Soc. 86(7):699–706. doi:10.1007/s11746-009-1376-5

Naes, T., T. Isaksson, T. Fearn, and T. Davies. 2002. A user friendly guide to multivariate calibration and classification. NIR publications, Chichester, UK.

Nassu, R. T., M. E. R. Dugan, M. L. He, T. A. McAllister, J. L. Aalhus, N. Aldai, and J. K. G. Kramer. 2011. The effects of feeding flaxseed to beef cows given forage based diets on fatty acids of longissimus thoracis muscle and backfat. Meat Sci. 89(4):469–477. doi:10.1016/j.meatsci.2011.05.016

Nuernberg, K., D. Dannenberger, G. Nuernberg, K. Ender, J. Voigt, N. D. Scollan, J. D. Wood, G. R. Nute, and R. I. Richardson. 2005. Effect of a grass-based and a concentrate feeding system on meat quality characteristics and fatty acid composition of longissimus muscle in different cattle breeds. Livest. Prod. Sci. 94(1–2):137–147. doi:10.1016/j.livprodsci.2004.11.036

Realini, C. E., Z. Kallas, M. Pérez-Juan, I. Gómez, J. L. Olleta, M. J. Beriain, P. Albertí, and C. Sañudo. 2014. Relative importance of cues underlying Spanish consumers' beef choice and segmentation, and consumer liking of beef enriched with n-3 and CLA fatty acids. Food Qual. Prefer. 33:74–85. doi:10.1016/j.foodqual.2013.11.007

Scollan, N., J. F. Hocquette, K. Nuernberg, D. Dannenberger, I. Richardson, and A. Moloney. 2006. Innovations in beef production systems that enhance the nutritional and health value of beef lipids and their relationship with meat quality. Meat Sci. 74(1):17–33. doi:10.1016/j.meatsci.2006.05.002

Scollan, N. D., D. Dannenberger, K. Nuernberg, I. Richardson, S. MacKintosh, J. F. Hocquette, and A. P. Moloney. 2014. Enhancing the nutritional and health value of beef lipids and their relationship with meat quality. Meat Sci. 97(3):384–394. doi:10.1016/j.meatsci.2014.02.015

Turner, T. D., J. L. Aalhus, C. Mapiye, D. C. Rolland, I. L. Larsen, J. A. Basarab, V. S. Baron, T. A. McAllister, H. C. Block, B. Uttaro, and M. E. R. Dugan. 2015. Effects of diets supplemented with sunflower or flax seeds on quality and fatty acid profile of hamburgers made with perirenal or subcutaneous fat. Meat Sci. 99:123–131. doi:10.1016/j.meatsci.2014.08.006

Vahmani, P., C. Mapiye, N. Prieto, D. C. Rolland, T. A. McAllister, J. L. Aalhus, and M. E. R. Dugan. 2015. The scope for manipulating the polyunsaturated fatty acid content of beef: A review. J. Anim. Sci. Biotechnol. 6(1):29. doi:10.1186/s40104-015-0026-z

Vahmani, P., D. C. Rolland, T. A. McAllister, H. C. Block, S. D. Proctor, L. L. Guand, N. Prieto, J. L. Aalhus, and M. E. R. Dugan. 2017. Effects of feeding steers extruded flaxseed mixed with hay or before hay on animal performance, carcass quality, and meat and hamburger fatty acid composition. Meat Sci. 131:9–17. doi:10.1016/j.meatsci.2017.04.008

Wong, E., L. N. Nixon, and C. B. Johnson. 1975. Volatile medium chain fatty acids and mutton flavor. J. Agric. Food Chem. 23(3):495–498. doi:10.1021/jf60199a044

Wood, J. D. 1990. Consequences for meat quality of reducing carcass fatness. In: J. D. Wood and A. V. Fisher, editors, Reducing fat in meat animals. Elsevier Applied Science, London. p. 344–397.

Wood, J. D., I. R. Richardson, G. R. Nute, A. V. Fisher, M. M. Campo, E. Kasapidou, P. R. Sheard, and M. Enser. 2004. Effects of fatty acids on meat quality: A review. Meat Sci. 66:21–32. doi:10.1016/S0309-1740(03)00022-6

Zhang, A., and Z. Y. Chen. 1997. Oxidative stability of conjugated linoleic acids relative to other polyunsaturated fatty acids. J. Am. Oil Chem. Soc. 74(12):1611–1613. doi:10.1007/s11746-997-0086-0