Archiv Tierzucht 56 (2013) 97, 958-970

Original study

Effect of breed and age on beef carcass quality, fatness and fatty acid composition

Nikolina Kelava Ugarković, Ante Ivanković and Miljenko Konjačić

Department of animal science and technology, Faculty of Agriculture, University of Zagreb, Zagreb, Croatia

Abstract

The aim of this research was to determine differences in beef carcass fatness and in the fatty acid composition of muscle and adipose tissue of three different breeds slaughtered at the age of 14 months and 19 months. The breed significantly affected the muscle fat content, carcass subcutaneous fat tissue thickness and fatty acid composition of the muscle and subcutaneous fat tissue. Different age at slaughter had no significant effect on analysed traits. The muscle tissue of the Herefords contained a higher ($P<0.05$) percentage of C14:0 and C16:0 fatty acids and fewer long-chain fatty acids than the Simmentals and Charolais. The subcutaneous fat tissue of the Simmentals contained a higher ($P<0.05$) percentage of PUFA, PUFA/SFA ratio and n-6 fatty acids. When fed with a high-energy diet, the Herefords proved to have the most fattened carcasses and the highest content of saturated fatty acids in the muscle. Changes in the diet of feedlot cattle should result in a more favourable fatty acid composition.

Keywords: beef, fat tissue, fatty acids, breed, age

Introduction

Beef is an important component of the human diet and is a source of valuable nutrients such as proteins, essential fatty acids, fat-soluble vitamins and minerals (Williamson *et al.* 2005, Nürnberg *et al.* 1999). However, lipids of beef meat are considered to have negative effect on human health. Beef is also considered as meat with a predominant percentage of saturated fatty acids responsible for an increase in LDL cholesterol (low-density lipoprotein) and for the development of coronary heart diseases. However, the predominant fatty acid

Archiv Tierzucht 56 (2013) 97, 958-970
doi: 10.7482/0003-9438-56-097

Received: 24 July 2013
Accepted: 6 November 2013
Online: 14 November 2013

Corresponding author:
Ante Ivanković; email: aivankovic@agr.hr
Department of Animal Science and Technology, Faculty of Agriculture University of Zagreb, Svetošimunska cesta 25, 10000 Zagreb, Croatia

in beef is oleic acid (C18:1 n-9c) and unsaturated fatty acids make up about 50 % of total fatty acids. Due to the low intramuscular fat percentage (2-5 %), beef can be considered to be low fat meat (Scollan *et al.* 2006, Nürnberg *et al.* 1999). The fat percentage and fatty acid composition can be affected by different factors, but genetic and nutritional factors are considered to contribute the most to differences in fatty acid composition (Scollan *et al.* 2006). Differences in the fatty acid composition between breeds occur due to a different gene expression or enzyme activity involved in fatty acid synthesis (de Smet *et al.* 2004). Among non-nutritional factors, age is an important factor affecting fatty acid composition. When the age progresses, the content of subcutaneous tissue and muscle fat increase, while the ratio of polyunsaturated (PUFA) vs. saturated (SFA) fatty acids declines (Warren *et al.* 2008). Consumers often have the perception that the meat of older cattle tends to be fattier and therefore has negative effects on human health.

Currently, the Simmental breed constitutes 61 % of all slaughtered cattle in Croatia, while the Hereford and Charolais make up 0.36 % and 0.17 %, respectively (CAA 2011). Croatia is known as an exporter of livestock and beef and with Italy as the most important export destination. The best known and most exported product is »baby beef«, meat of corn-fattened Simmental bulls (15-18 months) and heifers (12-14 months).

Based on the abovementioned facts, we can assume that the genotype and age of animals at slaughter have significant effects on the total intramuscular adipose tissue content and the muscle and subcutaneous fatty acid profiles. The objective of this research was to investigate the effects of genotype and different slaughter ages on carcass quality, fatness and the fatty acid composition of *musculus longissimus dorsi* (MLD) muscle and subcutaneous fat tissue.

Material and methods

The research included 175 steers randomly chosen from three different cattle breeds from the Croatian beef cattle population (60 Simmentals, 60 Herefords, 55 Charolais). The steers had been fattened in the same housing and feeding conditions, i.e. on a single farm, in different pens (approximately 10 steers per 60 m^2 pen). Steers of the Simmental, Hereford and Charolais breeds were fed *ad libitum* with the same diet, based on corn silage (46 %), wet corn (39 %), wheat straw (4.63 %) and concentrate with 34 % protein (10.37 %) distributed to the steers as total mixed ration twice a day. The average nutritional content of the feeds was approximately 7.6 MJ ME/kg dry matter and 950 g crude protein. The steers had *ad libitum* access to water during the whole fattening period. Eighty-eight animals (30 Simmentals, 30 Herefords, 28 Charolais) were slaughtered under controlled conditions in commercial abattoirs at the age of 14 months and 87 animals (30 Simmentals, 30 Herefords, 27 Charolais) at the age of 19 months. After 24 h *post-mortem*, the carcass EUROP conformation score and subcutaneous fat thickness were determined. Fat thickness was measured with a calliper over the 12th and 13th rib of the MLD at a point three-fourths of the length of the rib eye from the split chine bone (Tatum 2007). Approximately 50 g of subcutaneous fat tissue were taken at the 12th and 13th rib to determine subcutaneous fatty acid composition. To determine the total lipid percentage and fatty acid composition, approximately 100 g of the MLD were taken between the 12th and 13th rib. Muscle and fat samples were vacuum-packed individually and stored at –20 °C until analyses.

Crude fat determination

The total lipid content was determined gravimetrically by the Soxhlet method (ISO 1443:1973). After preparation, 5 g of a ground and homogenized meat sample were placed into a Soxhlet extractor and the extraction was carried out with n-hexane for 6 h with heating and solvent circulation of a minimum of 10 times per h. After extraction, the solvent was distilled and the residue was dried at 98 °C±2 °C for 1 h. It was left to cool in an exsiccator and weighed. It was then dried again for a further half an hour, left to cool and weighed. This was repeated until a difference between the last two measurements was less than 1 mg.

Analysis of fatty acid methyl esters

A sample quantity containing 1.0 g fat was destructed with 20 cm^3 of hydrochloric acid (37 %) for 1 h using a hot water bath. After being cooled down, 7 cm^3 of ethanol was added. Lipids were extracted with 15 cm^3 diethylether and 15 cm^3 benzine (b.p.<60 °C) and the organic layers were combined. From a portion of this solution containing 150 mg fat, the solvents were removed at 80 °C under reduced pressure. Next, 4 cm^3 of 0.5 M sodium hydroxide methanol solution were added to the residue and boiled until all the fat drops disappeared (5 min). Then, 4 cm^3 of 14 % boron trifluoride methanol solution were added and boiled for 3 min. Finally 4 cm^3 of hexane dried on water-free sodium sulphate were added and boiled for one minute and the mixture was allowed to cool. A saturated aqueous sodium chloride solution was added and, after being separated, the organic layer was collected into a 4 cm^3 vial containing water-free sodium sulphate and directly examined by gas chromatography.

Gas chromatographic analysis

Fatty acid methyl esters were quantified using a Shimadzu GC 2010 gas chromatograph equipped with a flame ionization detector and a CP-Sill 88 fused silica capillary column (100 m length, 0.25 mm wall coated open tubular-WCOT, 0.2 µm, Varian, USA). Analysis was performed using an initial temperature of 130 °C for 0 min and then the temperature was increased at a rate of 4 °C/min to 202 °C. At the end an isothermic period of 202 °C came for 15 min. The injector and detector were both maintained at 270 °C. Helium at 34.08 psi was used as the carrier gas. The sample injection split mode was 1:20. Fatty acids were identified by comparing the relative FAME (fatty acid methyl ester) peak retention times of the samples and the fatty methyl ester standards from Supelco (Supelco 37 Component Fame Mix 47885-U, Sigma-Aldrich). The PUFAs that were not included in this standard were determined by mass spectrometric detector. The peaks were determined by GCMS solution software (Shimadzu, UK). The fatty acids were expressed as percentages of each individual fatty acid to the total of all fatty acids present in the sample (Rowe *et al.* 1999).

Statistical analysis

Statistical analysis of the experimental data was performed using the General Linear Model procedure of SAS V9.2 (2008, SAS Institute Inc., Cary, NC, USA) including breed and age effects as independent variables. All tables contain the least square mean (LSMEAN) and standard error (SE) of the means. The level of significance of the treatment was set at $P<0.05$.

Results and discussion

The breed was a significant source of variation for fat content and subcutaneous fat tissue thickness (Table 1). The muscle fat content and subcutaneous fat thickness were greater (P<0.001 i.e. 0.05) in Herefords than in Simmentals and Charolais (3.15 %, 0.69 cm vs. 1.79, 0.46; 1.28, 0.34, respectively). The EUROP conformation and carcass fat classification showed no significant variation between breeds. According to the breed's physiology, the Charolais had the highest conformation scores and the Hereford the greatest fat classification scores. The higher fatness of the Herefords can be explained by the high energy diet. The Herefords, due to the breed´s reproductive (early maturing) and physiological characteristics, when fed with a high energy diet (grain-finished), yield more fattened carcasses. Other authors (Bureš *et al.* 2006, Realini *et al.* 2004) have reported on similar results for the Hereford. Higher muscle tissue fat percentage for Simmental was found by several authors (Laborde *et al.* 2001, Nuernberg *et al.* 2005, Mahecha *et al.* 2009). Since animals were kept and fed under comparable conditions during the experiment, it can be suggested that their differences in muscle fat content and fatty acid composition are influenced by the breed.

No significant association was found between the age at slaughter and the analysed carcass characteristics (Table 1). Somewhat higher values of the majority of the analysed carcass traits were found at the age of 19 months. The muscle fat content showed the greatest difference in the different ages of the slaughter groups (2.34 vs. 1.81 %). Lengyel *et al.* (2003) also reported on no significant difference in the intramuscular fat percentage between the 14 and 19 month age groups. Although some authors (Martin *et al.* 1999, Robelin 1986) observed an increase in the fat percentage with age, it seems that as age progresses differences in the fat percentage diminishes. While the intramuscular depot is the last developing fat depot, greater differences are between young animals with less intramuscular fat tissue and older animals with more intramuscular fat tissue. The differences between older animals are mainly due to the different percentage of neutral lipids in the intramuscular fat tissue (Lengyel *et al.* 2003).

Table 1
Carcass characteristics of the analyzed breeds slaughtered at different ages (LSMEAN±SE)

Trait	Breed			Slaughter age, months		Significance	
	Simmental	Hereford	Charolais	14	19	breed	age
Dry matter in MLD, %	25.30±0.33[a]	26.85±0.33[b]	24.63±0.42[a]	25.43±0.34	25.75±0.34	*	ns
Fat in MLD, %	1.79±0.27[a]	3.15±0.26[b]	1.28±0.33[b]	1.81±0.28	2.34±0.28	**	ns
Subcutaneous fat tissue, cm	0.46±0.10[ab]	0.69±0.10[a]	0.34±0.10[b]	0.48±0.06	0.51±0.06	*	ns
EUROP conformation[a]	4.32±0.18	4.22 ±0.18	4.69±0.23	4.48±0.15	4.27±0.15	ns	ns
EUROP fat classification[b]	3.24±0.17	3.35±0.17	3.04±0.21	3.15±0.13	3.27±0.13	ns	ns

[a]conformation: 1=poorest, 5=excellent; [b]fat classification: 1=leanest, 5=fattest; MLD: *musculus longissimus dorsi*; ns: not significant; *P<0.05, **P<0.001

The fatty acid composition of muscle tissue differed significantly between breeds (Table 2). The percentage of odd-chain fatty acids was significantly lower in the Simmental than in the Hereford and Charolais. The Hereford had a significantly higher C14:0, C16:0 and C16:1 percentage than the Simmental and Charolais. The percentage of oleic acid (C18:1 n-9c) was significantly lower in the Charolais than in the Simmental and Hereford. In the Simmental and Hereford muscle tissue, significantly higher percentage of conjugated linoleic acid

(CLA) and C20:1 was found than in the Charolais. In the Hereford muscle tissue, significantly lower percentage of long-chain PUFAs was found. A significantly lower SFA content was found in the Simmental compared to the Hereford and Charolais. Although the highest SFA percentage was detected in the Charolais, this was due to the highest C18:0 percentage. While significantly higher C16:0 contents and the highest C14:0 percentage was found in the Hereford, this breed had the least favourable SFAs regarding the effect on human health. Graham *et al.* (2006) reported on significantly less SFAs in the Simmental compared with Hereford.

Table 2
Fatty acid composition (% of total fatty acids) in MLD of analyzed breeds (LSMEAN±SE)

Fatty acids	Breeds			Significance
	Simmental	Hereford	Charolais	
C10:0	0.044±0.01	0.054±0.01	0.051±0.01	ns
C12:0	0.055±0.01	0.059±0.01	0.069±0.01	ns
C14:0	2.168±0.24^{a}	3.134±0.26^{b}	2.671±0.29^{ab}	*
C14:1	0.350±0.07	0.576±0.07	0.322±0.08	ns
C15:0	0.320±0.03^{a}	0.452±0.03^{b}	0.466±0.03^{b}	*
C16:0	24.785±0.72^{a}	28.116±0.80^{b}	25.799±0.87^{ab}	*
C16:1	3.049±0.24^{ab}	3.832±0.27^{a}	2.666±0.29^{b}	*
C17:0	0.928±0.07^{a}	1.339±0.08^{b}	1.136±0.08^{ab}	*
C18:0	18.996±0.91	17.648±1.01	20.910±1.09	ns
C18:1n-9t	2.713±0.34	2.582±0.37	2.246±0.40	ns
C18:1n-9c	36.363±1.34^{a}	37.909±1.48^{a}	31.033±1.60^{b}	*
C18:2n-6	4.743±0.89	2.680±0.99	5.584±1.07	ns
C18:2c9.t11	0.366±0.03^{a}	0.316±0.03^{a}	0.185±0.03^{b}	*
C18:3n-3	0.172±0.02	0.168±0.02	0.146±0.02	ns
C20:0	0.133±0.01	0.100±0.01	0.145±0.01	ns
C20:1	0.142±0.01^{a}	0.138±0.01^{a}	0.105±0.01^{b}	*
C20:2	0.104±0.03	0.044±0.03	0.079±0.03	ns
C20:3n-6	0.551±0.09^{a}	0.132±0.10^{b}	0.674±0.11^{a}	*
C20:4n-6	1.204±0.24^{a}	0.327±0.26^{a}	1.882±0.28^{b}	*
C20:5n-3	0.136±0.02^{a}	0.026±0.03^{b}	0.174±0.03^{a}	*
C22:5n-3	0.175±0.04^{ab}	0.103±0.04^{a}	0.285±0.04^{b}	*
SFA[1]	47.429±0.73^{a}	50.902±0.81^{b}	51.248±0.87^{b}	**
MUFA[2]	42.617±1.52^{a}	45.037±1.69^{a}	36.373±1.83^{b}	*
PUFA[3]	9.616±1.54^{a}	3.546±1.71^{b}	12.133±1.85^{a}	*
n-6[4]	6.498±1.18	3.139±1.31	8.139±1.42	ns
n-3[5]	0.483±0.07^{ab}	0.297±0.07^{a}	0.606±0.08^{b}	*
PUFA/SFA[6]	0.203±0.03^{a}	0.07±0.03^{b}	0.238±0.07^{a}	*
n-6/n-3[7]	13.593±1.13	10.292±1.26	13.582±1.36^{ab}	ns
Δ^{9}-desaturase (16) index	10.900±0.58^{ab}	12.055±0.64^{a}	9.285±0.69^{b}	*
Δ^{9}-desaturase (18) index	67.165±1.75^{ab}	69.767±1.94^{a}	61.389±2.10^{b}	*

[1]sum of saturated fatty acids, [2]sum of monounsaturated fatty acids, [3]sum of polyunsaturated fatty acids + unidentified PUFA, [4,5]sum of n-6 and n-3 fatty acids, [6]ratio of the sum of PUFA and SFA, [7]ratio of the sum of n-6 and n-3 fatty acids, [8]Δ^{9}-desaturase (16) index=100(16:1n-9/(16:1n-9+16:0)), [9]Δ^{9}-desaturase (18) index=100(18:1n-9/(18:1n-9+18:0)), ns: not significant, *P<0.05, **P<0.001

A significantly higher monounsaturated (MUFA) fatty acids percentage was found in the Herford and Simmental in comparison with the Charolais. Monounsaturated fatty acids have a neutral effect on the human blood cholesterol level because predominant oleic acid is a favoured substrate for the liver enzyme that converts cholesterol to an inactive form (Whetsell *et al.* 2003, Grundy 1994). Other MUFAs also have a favourable effect on human health due to their transformation into beneficial long-chain fatty acids (Whetsell *et al.* 2003, Burdge & Wootton 2002). A significantly lower level of n-3 fatty acids was found in the Hereford compared with the Charolais. Due to the lowest n-6 fatty acids percentage in the Hereford there was also found the lowest n-6/n-3 ratio. Total PUFAs in Hereford were found to be 41.87 % and 34.58 % lower than in the Charolais and Simmental.

Table 3
Fatty acid composition (% of total fatty acids) in MLD of steers slaughtered at different age (LSMEAN±SE)

Fatty acids	Slaughter age (months)		Significance
	14	19	
C10:0	0.051±0.02	0.048±0.01	0.544
C12:0	0.061±0.01	0.060±0.02	0.940
C14:0	2.630±0.54	2.685±0.64	0.830
C14:1	0.399±0.15	0.433±0.18	0.643
C15:0	0.433±0.07	0.393±0.11	0.169
C16:0	26.003±1.78	26.463±2.03	0.575
C16:1	3.232±0.78	3.132±0.49	0.719
C17:0	1.162±0.18	1.108±0.22	0.504
C18:0	18.898±2.23	19.471±2.17	0.544
C18:1n-9t	2.642±1.01	2.386±0.57	0.432
C18:1n-9c	33.954±4.05	36.249±3.18	0.121
C18:2n-6	5.099±2.52	3.571±1.46	0.114
C18:2c9,t11	0.305±0.10	0.273±0.11	0.231
C18:3n-3	0.177±0.11	0.147±0.07	0.061
C20:0	0.133±0.04	0.120±0.02	0.364
C20:1	0.126±0.05	0.131±0.02	0.701
C20:2	0.079±0.05	0.072±0.07	0.809
C20:3n-6	0.527±0.30	0.378±0.19	0.198
C20:4n-6	1.287±0.69	0.988±0.61	0.313
C20:5n-3	0.132±0.08	0.092±0.05	0.197
C22:5n-3	0.219±0.13	0.157±0.07	0.129
SFA[1]	49.372±4.48	50.348±4.16	0.196
MUFA[2]	40.353±4.26	42.332±4.02	0.249
PUFA[3]	9.827±0.68	7.035±0.32	0.143
n-6[4]	6.914±3.40	4.937±2.17	0.139
n-3[5]	0.528±0.28	0.396±0.13	0.088
PUFA/SFA[6]	0.200±0.02	0.141±0.01	0.131
n-6/n-3[7]	12.847±3.20	12.130±3.65	0.551
Δ^9-desaturase (16) index[8]	10.953±0.43	10.541±0.44	0.514
Δ^9-desaturase (18) index[9]	65.764±1.30	66.450±1.29	0.715

[1]sum of saturated fatty acids, [2]sum of monounsaturated fatty acids, [3]sum of polyunsaturated fatty acids, [4,5]sum of n-6 and n-3 fatty acids, [6]ratio of the sum of PUFA and SFA, [7]ratio of the sum of n-6 and n-3 fatty acids, [8]Δ^9-desaturase (16) index=100(16:1n-9/(16:1n-9+16:0)), [9]Δ^9-desaturase (18) index=100(18:1n-9/(18:1n-9+18:0))

The age at slaughter showed no association with the fatty acid composition in the muscle tissue (Table 3). At the age of 19 months, a significantly higher C18:1 n-9c percentage was detected in the muscle tissue than at the age of 14 months. Higher percentages of linoleic, α-linolenic, C20:3 n-6 and C22:5 n-3 fatty acids were found in the muscle tissue at the age of 14 months. Due to the percentage of individual fatty acids, at the age of 14 months higher PUFA, n-6 fatty acids percentages and polyunsaturated/saturated fatty acids (PUFA/SFA) ratio were found. These results correspond to the known dynamics of changes in muscle tissue. With an increase in age, the percentage of neutral lipids increases in the muscle and consequently the percentages of phospholipids and PUFAs decrease (Eichhorn *et al.* 1985 cit. Lengyel *et al.* 2003, Malau-Aduli *et al.* 1998). Similar to the results of earlier researches (Martin *et al.* 1999, Robelin 1986), it can be concluded that in the muscle tissue specific changes in fatty acid composition occur with age, noticeably an increase of saturated and monounsaturated fatty acids. However, with age the percentage of total polyunsaturated, n-6 and n-3 fatty acids decreases.

Analysing the indices of desaturase enzyme activity in the muscle tissue, it was found that the Hereford MLD had higher ($P<0.05$) values of Δ^9 desaturase (16) and (18) than the Charolais. Higher values of both Δ^9 desaturase enzymes in the Hereford were reported by other authors (Siebert *et al.* 2003, Garcia *et al.* 2007). As Δ^9 desaturase enzymes convert C16:0 to C16:1 and C18:0 to C18:1 in conditions of a grain-based diet, the Hereford seems to have a genetic basis for pronounced Δ^9 desaturase activity (Garcia *et al.* 2007). A significant difference of the Δ^9 desaturase (16) and (18) activity was not found in the muscle tissue in different age groups or in the subcutaneous fat tissue of different breeds and age groups.

The breed showed a significant association with the subcutaneous fatty acid composition (Table 4). In the subcutaneous fat tissue of the Simmental, a significantly lower percentage of odd-chain fatty acids was found than in the Hereford or Charolais. In addition, in the subcutaneous fat tissue of the Simmental, a significantly higher C18:2 n-6 percentage was found than in the Hereford and Charolais. Due to the individual fatty acid percentage, in the subcutaneous fatty acids of the Simmental significantly higher PUFAs and a n-6 fatty acid percentage, as well as PUFA/SFA ratio, were found than in the Hereford or Charolais. A lower percentage of palmitic and stearic fatty acids was found in the Simmental subcutaneous fat tissue, while the Hereford tended to have a more unfavourable percentage of these saturated fatty acids. Although the Hereford had a significantly lower percentage of PUFAs and n-6 fatty acids in its subcutaneous fat tissue, the n-6/n-3 ratio of the Hereford subcutaneous fat tissue had the lowest value. Subcutaneous fat tissue constitutes up to 17 % of the total fat tissue in beef carcasses (Nürnberg *et al.* 1998). During the standard processing of carcasses on the slaughter line excess subcutaneous fat tissue is cut down, that is why subcutaneous fat tissue contributes less to the total fat percentage than intermuscular fat tissue. The subcutaneous fat tissue of the analysed breeds had on average less saturated and polyunsaturated fatty acids than muscle tissue, while the average MUFA in the subcutaneous fat tissue was higher than in the total lipids of the muscle tissue. The average PUFA/SFA and n-6/n-3 ratios in the subcutaneous fat tissue of the analysed breeds were less favourable than in the muscle tissue.

Table 4
Fatty acid composition (% of total fatty acids) of subcutaneous fat tissue of analyzed breeds (LSMEAN±SE)

Fatty acids	Breeds			Significance
	Simmental	Hereford	Charolais	
C10:0	0.037±0.01	0.054±0.01	0.054±0.01	ns
C12:0	0.065±0.01	0.071±0.01	0.098±0.01	ns
C14:0	3.247±0.29	3.785±0.29	3.970±0.32	ns
C14:1	0.810±0.15	1.218±0.16	0.983±0.17	ns
C15:0	0.439±0.05[a]	0.674±0.05[b]	0.704±0.05[b]	*
C16:0	26.247±0.89	28.211±0.91	26.880±0.99	ns
C16:1	5.130±0.36	4.555±0.37	5.211±0.40	ns
C17:0	0.985±0.11[a]	1.594±0.11[b]	1.267±0.12[ab]	*
C18:0	14.808±1.22	14.947±1.24	15.535±1.35	ns
C18:1n-9t	3.883±0.41	3.732±0.41	3.091±0.44	ns
C18:1n-9c	40.453±1.54	38.457±1.57	39.191±1.70	ns
C18:2n-6	2.609±0.16[a]	1.743±0.16[b]	1.979±0.18[b]	*
C18:2c9,t11	0.142±0.01	0.160±0.02	0.126±0.02	ns
C18:3n-3	0.187±0.03	0.152±0.03	0.163±0.03	ns
C20:0	0.088±0.01	0.088±0.01	0.127±0.01	ns
C20:1	0.100±0.01	0.103±0.01	0.099±0.01	ns
C20:2	0.026±0.01	0.023±0.01	0.031±0.01	ns
C21:0	0.646±0.06[a]	0.362±0.06[b]	0.397±0.07[b]	*
C20:3n-6	0.050±0.01	0.051±0.01	0.043±0.01	ns
C20:4n-6	0.036±0.01	0.029±0.01	0.058±0.04	ns
SFA[1]	46.562±1.77	49.784±1.80	49.031±1.96	ns
MUFA[2]	50.375±1.75	48.064±1.78	48.574±1.93	ns
PUFA[3]	3.050±0.18[a]	2.158±0.19[b]	2.400±0.20[ab]	**
n-6[4]	2.695±0.17[a]	1.824±0.17[b]	2.080±0.18[b]	*
n-3[5]	0.187±0.03	0.152±0.03	0.163±0.03	ns
PUFA/SFA[6]	0.066±0.01[a]	0.043±0.01[b]	0.049±0.01[ab]	*
n-6/n-3[7]	18.042±3.04	12.767±3.09	14.881±3.35	ns
Δ^9-desaturase (16) index[8]	16.387±1.03	13.911±1.05	16.247±1.13	ns
Δ^9-desaturase (18) index[9]	74.814±2.04	73.892±2.07	73.125±2.25	ns

[1]sum of saturated fatty acids, [2]sum of monounsaturated fatty acids, [3]sum of polyunsaturated fatty acids, [4,5]sum of n-6 and n-3 fatty acids, [6]quotient of the sum of PUFA and SFA, [7]quotient of the sum of n-6 and n-3 fatty acids, [8]Δ^9-desaturase (16) index=100(16:1n-9/(16:1n-9+16:0)), [9]Δ9-desaturase (18) index=100(18:1n-9/(18:1n-9+18:0)), ns: not significant, *P<0.05, **P<0.01, ***P<0.0001

The age at slaughter showed no significant association with the subcutaneous fatty acid composition (Table 5). Higher percentages of predominant SFAs (C14:0, C16:0, C18:0) were found at the age of 14 months. At the age of 19 months higher percentages of C18:1 n-9c and C18:3 n-3 fatty acids were found in the subcutaneous fat tissue and a lower percentage of CLA. The percentage of other PUFAs decreased with age. At the age of 14 months, 30 % more C14:0 was found in the subcutaneous fat tissue than in the muscle tissue, while at 19 months the difference was 23 %. Contrary to Scollan *et al.* (2001) a higher percentage of palmitic and stearic fatty acids was found in the subcutaneous fat tissue than in the muscle tissue. For both age groups, more favourable PUFA/SFA and n-6/n-3 ratios were found in the muscle tissue than in the subcutaneous fat tissue.

Table 5
Fatty acid composition (% of total fatty acids) of subcutaneous fat tissue steers slaughtered at different age (LSMEAN±SE)

Fatty acids	Slaughter age (months)		Significance
	14	19	
C10:0	0.049±0.01	0.047±0.01	0.733
C12:0	0.081±0.02	0.074±0.03	0.449
C14:0	3.852±0.68	3.482±0.71	0.166
C14:1	0.933±0.35	1.073±0.32	0.348
C15:0	0.651±0.13	0.559±0.17	0.062
C16:0	27.587±2.25	26.638±1.20	0.273
C16:1	4.912±0.68	5.019±0.96	0.753
C17:0	1.321±0.34	1.243±0.39	0.523
C18:0	15.486±2.80	14.707±2.80	0.486
C18:1n-9t	3.677±0.89	3.460±0.89	0.556
C18:1n-9c	38.112±4.20	40.621±3.27	0.088
C18:2n-6	2.217±0.63	2.004±0.28	0.214
C18:2c9,t11	0.147±0.06	0.138±0.08	0.525
C18:3n-3	0.159±0.07	0.175±0.06	0.554
C20:0	0.101±0.02	0.101±0.03	0.965
C20:1	0.097±0.02	0.104±0.02	0.236
C20:2	0.032±0.02	0.021±0.01	0.094
C21:0	0.482±0.25	0.454±0.19	0.639
C20:3n-6	0.051±0.03	0.044±0.02	0.424
C20:4n-6	0.052±0.03	0.030±0.02	0.070
SFA[1]	49.611±4.99	47.307±3.71	0.171
MUFA[2]	47.732±4.77	50.278±3.61	0.125
PUFA[3]	2.659±0.68	2.414±0.31	0.203
n-6[4]	2.321±0.64	2.079±0.29	0.179
n-3[5]	0.159±0.07	0.175±0.06	0.554
PUFA/SFA[6]	0.054±0.02	0.051±0.01	0.539
n-6/n-3[7]	16.593±7.83	13.867±6.28	0.339
Δ^9-desaturase (16) index[8]	15.169±0.70	15.861±0.69	0.494
Δ^9-desaturase (18) index[9]	72.893±1.28	74.995±1.25	0.264

[1]sum of saturated fatty acids, [2]sum of monounsaturated fatty acids, [3]sum of polyunsaturated fatty acids, [4,5]sum of n-6 and n-3 fatty acids, [6]quotient of the sum of PUFA and SFA, [7]quotient of the sum of n-6 and n-3 fatty acids, [8]Δ^9-desaturase (16) index=100(16:1n-9/(16:1n-9+16:0)), [9]Δ^9-desaturase (18) index=100(18:1n-9/(18:1n-9+18:0))

Changes in the fatty acid composition between the MLD and subcutaneous fat tissue were significant across breeds and different slaughter age groups (Table 6). A significantly higher percentage of C12:0 was found in the subcutaneous fat tissue at the age of 14 months than in the MLD. Across all breeds and both slaughter age groups, significantly higher percentages of fatty acids C14:0, C14:1, C15:0, C16:1 were found in the subcutaneous fat tissue than in the MLD. The Herefords' subcutaneous fat tissue had a significantly higher percentage of C17:0 than the MLD. A significantly higher percentage of C18:0 was found in the MLD across all breeds and in both slaughter age groups than in the subcutaneous fat tissue. Across both slaughter age groups, a significantly lower percentage of C18:1 n-9t was found in the MLD than in the subcutaneous fat tissue. Both slaughter age groups and the Charolais subcutaneous fat tissue were found to have a higher percentage of C18:1 n-9c fatty acid than the MLD. The *musculus longissimus dorsi* of the Charolais and the younger slaughter age group were found to have

a significantly higher percentage of C18:2 n-6 than the subcutaneous fat tissue. Across all breeds and both slaughter age groups, a significantly higher percentage of CLA, C20:3 n-6 and C20:4 n-6 was found in the MLD than in the subcutaneous fat tissue. Both slaughter age groups as well as the Simmentals' and Herefords' MLD were found to have a significantly higher percentage of C20:1 than the subcutaneous fat tissue. A significantly lower percentage of C20:2 was found in the subcutaneous fat tissue at the age of 19 months than in the MLD. In the MLD of the Simmental and Charolais in both age groups, significantly higher percentages of PUFAs, n-6, n-3 fatty acids and PUFA/SFA ratio were found than in the subcutaneous fat tissue. In the MLD of the Simmentals and Herefords in both age groups, a significantly higher MUFA percentage was found than in the subcutaneous fat tissue.

Table 6
Differences in fatty acid composition (% of total fatty acids) of MLD and subcutaneous fat tissue of steers slaughtered at different age

Fatty acids	Tissue (LSMEAN)		SE	Significance
	MLD	SUBCUT		
C10:0	0.049	0.048	0.003	ns
C12:0	0.061	0.078	0.004	A14
C14:0	2.658	3.667	0.13	B, A
C14:1	0.416	1.003	0.05	B, A
C15:0	0.413	0.605	0.03	B, A
C16:0	26.233	27.113	0.42	ns
C16:1	3.182	4.965	0.14	B, A
C17:0	1.135	1.282	0.05	BH
C18:0	19.185	15.097	0.48	B, A
C18:1n-9t	2.514	3.569	0.17	A
C18:1n-9c	35.102	39.367	0.76	BC, A
C18:2n-6	4.335	2.110	0.33	BC, A14
C18:2c9,t11	0.289	0.143	0.01	B, A
C18:3n-3	0.162	0.167	0.01	ns
C20:0	0.126	0.101	0.01	ns
C20:1	0.129	0.100	0.01	BS,H, A
C20:2	0.076	0.027	0.01	A19
C20:3n-6	0.453	0.048	0.04	B, A
C20:4n-6	1.138	0.041	0.10	B, A
SFA[1]	49.859	48.459	0.68	ns
MUFA[2]	41.343	49.005	0.86	BS,H, A
PUFA[3]	8.432	2.536	0.63	BS,C, A
n-6[4]	5.925	2.199	0.01	BS,C ;A
n-3[5]	0.462	0.167	0.45	BS,C, A
PUFA/SFA[6]	0.170	0.053	0.03	BS, C; A
n-6/n-3[7]	12.489	15.230	1.14	ns
Δ^9-desaturase (16) index[8]	10.747	15.515	0.41	B, A
Δ^9-desaturase (18) index[9]	64.525	72.228	0.96	B, A

MLD: *musculus longissimus dorsi*, SUBCUT: subcutaneous fat tissue, [1]sum of saturated fatty acids, [2]sum of monounsaturated fatty acids, [3]sum of polyunsaturated fatty acids, [4,5]sum of n-6 and n-3 fatty acids, [6]quotient of the sum of PUFA and SFA, [7]quotient of the sum of n-6 and n-3 fatty acids, [8]Δ^9-desaturase (16) index=100(16:1n-9/(16:1n-9+16:0)), [9]Δ^9-desaturase (18) index=100(18:1n-9/(18:1n-9+18:0)), ns: not significant, B, BS, BH, BC: significant effect of all breeds, Simmental, Hereford, Charolais, A, A14: significant effect of both age groups, age group 14 months

Although nutrition is considered to be a major factor influencing the fatty acid composition of beef (Scollan *et al.* 2006), due to the same keeping and feeding conditions in this study, it was shown that differences in fatty acid composition are influenced by genetic factors i.e. by the breed. These results correspond to the results reported by other authors (Huerta-Leidenz *et al.* 1993, Zembayashi *et al.* 1995, Malau-Aduli *et al.* 1998, Nürnberg *et al.* 1999). The greatest fat content and the thickest subcutaneous fat tissue were found in the Hereford. This is the result of the genetic and physiological characteristics of the Hereford breed which, with a concentrate diet, results in more fattened carcasses. Changes in the Herefords' diet could be recommended, especially by introducing a forage and pasture based diet that results in lower muscle fat content, higher PUFAs and better PUFA/SFA ratios (Realini *et al.* 2004, Nuernberg *et al.* 2005, Scollan *et al.* 2006, Warren *et al.* 2008). The research showed no significant differences in carcass fat characteristics as the slaughter age increased. Šubrt *et al.* (2006) reported that factors such as the age of the animals and the carcass weight had no effect on the structure of fatty acids. The major difference was found in the muscle fat content that increased with age, mainly due to the increase of neutral lipids. The content of muscle unsaturated fatty acids changed with an increase in age.

The discussion of the fatty acid composition of beef and the effect on human health has been relatively controversial (Nuernberg *et al.* 2005). Controversy arose from drawing attention to beef SFAs and cholesterol related to a higher risk of coronary heart disease (Pfeuffer 2001). However, only palmitic and miristic fatty acids as dominant saturated fatty acids are responsible for a rise in LDL. Lean beef meat contains favourable contents of MUFAs, PUFAs and other nutrients, and should be included as a component of a healthy balanced diet. The PUFA/SFA and n-6/n-3 ratios of muscle and subcutaneous fat tissue for all analysed breeds deviated from dietary recommendations.

In Croatia, due to grain and corn-based diets during beef fattening, certain genotypes show excessive carcass fatness and less favourable fatty acid compositions and ratios. Due to the growth characteristics and physiology of different breeds, the Hereford was found to be the least profitable breed in these fattening conditions. In order to obtain less fattened carcasses and meat with less SFA changes during fattening, technology should be adapted to the specific genotype requirements.

In the present study, the breed was found to have a significant effect on muscle and subcutaneous fat tissue. A less favourable subcutaneous fat thickness and fatty acid composition was especially noted for Hereford carcasses, where the muscle tissue contained higher proportion of SFAs and a lower proportion of PUFAs than the other analysed breeds. The age at slaughter showed no significant effect on the analysed traits in these experimental conditions. Considering the observed carcass traits, fatty acid proportions and ratios, a change of feeding strategy, especially for Herefords, can be recommended in order to enhance the nutritional value of beef according to dietary recommendations.

Acknowledgements

This research was supported by the Ministry of Science, Education and Sports, Croatia (Project 178-0790466-0398).

References

Bureš D, Bartoň L, Zahrádková R, Teslík V, Krejčová M (2006) Chemical composition, sensory characteristics, and fatty acid profile of muscle from Aberdeen Angus, Charolais, Simmental and Hereford bulls. Czech J Anim Sci 51, 279-284

CAA – Croatian Agriculture Agency (2011) Annual report. Zagreb, Croatia (available: http://www.hpa.hr)

De Smet S, Raes K, Demeyer D (2004) Meat fatty acid composition as affected by fatness and genetic factors: a review. Anim Res 53, 81-98

Eichhorn JM, Bailey CM, Blomquist GJ (1985) Fatty Acid Composition of Muscle and Adipose Tissue from Crossbred Bulls and Steers. J Anim Sci 61, 892-904

Garcia PT, Pensel NA, Sancho AM, Latimori NJ, Kloster AM, Amigone MA, Casal JJ (2007) Beef lipids in relation to animal breed and nutrition in Argentina. Meat Sci 79, 500-508

Graham JF, Bernaud E, Deland MPB (2006) Sire and dam breed effects on fatty acid profiles in the longissimus dorsi muscle and subcutaneous fat of beef cattle. Aust J Exp Agr 46, 913-919

Grundy SM (1994) Influence of stearic acid on cholesterol metabolism relative to other long-chain fatty acids. Am J Clin Nutr 60, 986S-990S

Huerta-Leidenz NO, Cross HR, Savell JW, Lunt DK, Baker JF, Pelton LS, Smith SB (1993) Comparison of the fatty acid composition of subcutaneous adipose tissue from mature Brahman and Hereford cows. J Anim Sci 71, 625-630

ISO (International Organization for Standardization) (1973) Determination of total fat content, ISO 1443:1973 standard. In: International standards meat and meat products. International Organization for Standardization, Genéve, Switzerland

Laborde FL, Mandell IB, Tosh JJ, Wilton JW, Buchanan-Smith JG (2001) Breed effects on growth performance, carcass characteristics, fatty acid composition, and palatability attributes in finishing steers. J Anim Sci 79, 355-365

Lengyel Z, Husvéth F, Polgár P, Szabó F, Magyar L (2003) Fatty acid composition of intramuscular lipids in various muscles of Holstein-Friesian bulls slaughtered at different ages. Meat Sci 65, 593-598

Mahecha L, Nuernberg K, Nuernberg G, Ender K, Hagemann E, Dannenberger D (2009) Effects of diet and storage on fatty acid profile, micronutrients and quality of muscle from German Simmental bulls. Meat Sci 82, 365-371

Malau-Aduli AE, Siebert BD, Bottema CD, Pitchford WS (1998) Breed comparison of the fatty acid of muscle phospholipids in Jersey and Limousin cattle. J Anim Sci 76, 766-773

Martin GS, Lunt DK, Britain KG, Smith SB (1999) Postnatal development of stearoyl coenzyme A desaturase gene expression and adiposity in bovine subcutaneous adipose tissue. J Anim Sci 77, 630-636

Nuernberg K, Dannenberger D, Nuernberg G, Ender K, Voigt J, Scollan ND, Wood JD, Nute GR, Richardson RI (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, 137-147

Nürnberg K, Ender B, Papstein HJ, Wegner J, Ender K, Nürnberg G (1999) Effects of growth and breed on the fatty acid composition of the muscle lipids in cattle. Zeitschr Lebensm Unters Forsch A 208, 332-335

Nürnberg K, Wegner J, Ender K (1998) Factors influencing fat composition in muscle and adipose tissue of farm animals. Livest Prod Sci 56, 145-156

Pfeuffer M (2001) Physiologic effects of individual fatty acids in animal and human body, with particular attention to coronary heart disease risk modulation. Arch Tierz 44, 89-98

Realini CE, Duckett SK, Brito GW, Dalla Rizza M, De Mattos D (2004) Effect of pasture vs. concentrate feeding with or without antioxidants on carcass characteristics, fatty acid composition, and quality of Uruguayan beef. Meat Sci 66, 567-577

Robelin J (1986) Growth and adipose tissues in cattle; partitioning between depots, chemical composition and cellularity. A review. Livest Prod Sci 14, 349-364

Rowe A, Macedo FAF, Visentainer JV, Souza NE, Matsuhita M (1999) Muscle composition and fatty acid profile in lambs fattened in drylot or pasture. Meat Sci 54, 238-288

SAS (2008) The Statistical Analysis System, v9.2. SAS Institute Inc., Cary, NC, USA

Scollan N, Hocquette JF, Nuernberg K, Dannenberger D, Richardson I, Moloney A (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, 17-33

Scollan ND, Choi NJ, Kurt E, Fisher AV, Enser M, Wood JD (2001) Manipulating the fatty acid composition of muscle and adipose tissue in beef cattle. Brit J Nutr 85, 115-124

Siebert BD, Pitchford WS, Kruk ZA, Kuchel H, Deland MPB, Bottema CDK (2003) Differences in Δ9 desaturase activity between Jersey- and Limousin-sired cattle. Lipids 38, 539-543

Šubrt J, Filipčík R, Župka Z, Fialová M, Dračková E (2006) The content of polyunsaturated fatty acids in intramuscular fat of beef cattle in different breeds and crossbreeds. Arch Tierz 49, 340-350

Tatum G (2007) Beef grading. Beef Facts. Product Enhancement. National Cattlemen´s Beef Association, Centennial, CO, USA http://www.beefresearch.org/CMDocs/BeefResearch/Beef%20Grading.PDF (last accessed 20.01.2014)

Warren HE, Scollan ND, Enser M, Hughes SI, Richardson RI, Wood JD (2008) Effects of breed and a concentrate or grass silage diet on beef quality in cattle of 3 ages. I: Animal performance, carcass quality and muscle fatty acid composition. Meat Sci 78, 256-269

Whetsell MS, Rayburn EB, Lozier JD (2003) Human Health Effects of Fatty Acids in Beef. A report on »Pasture based beef systems for Appalachia,« West Virginia University, Morgantown, WV, USA http://www.wvu.edu/~agexten/forglvst/humanhealth.pdf (last accessed 20.01.2014)

Williamson CS, Foster RK, Stanner SA, Buttriss JL (2005) Red meat in the diet. Nutr Bull 30, 323-355

Zembayashi M, Nishimura K, Lunt DK, Smith SB (1995) Effect of breed type and sex on the fatty acid composition of subcutaneous and intramuscular lipids of finishing steers and heifers. J Anim Sci 73, 3325-3332