FAT QUALITY IN THE INDIGENOUS KRŠKOPOLJE PIG REARED IN AN ENRICHED ENVIRONMENT

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The search for fat composition and content that is favourable in intramuscular fat (IMF) for fresh meat quality and at the same time in subcutaneous adipose tissue (SCF) for processed meat products, within the same breed, is reflected in the growing interest in native pig breeds. The aim of this study was to determine the fatty acid composition of the IMF of M. longissimus dorsi (LD) and adjacent SCF from the Slovenian indigenous Krškopolje pig. The type of fat from the 42 Krškopolje pig significantly affected the proportions of 29 of the 35 fatty acids measured here. The SCF contained higher proportions of saturated fatty acids (SFA) and polyunsaturated fatty acids (PUFA) and correspondingly less monounsaturated fatty acids (MUFA) than the IMF. Taking the extremes, the IMF at 6-10 % of the LD had higher proportions of SFA and MUFA and less PUFA than the IMF at 2-3 %. The thicker SCF (57-67 mm) was higher in SFA and lower in PUFA than thinner (30-40 mm). Sex and age also affected fatty acid composition; the fat from barrows was higher in MUFA and lower in PUFA than that from gilts, and with increasing age, MUFA content increased.

Key words: pigs / autochthonous breeds / Krškopolje pig / enriched environment / fatty acid composition / intramuscular fat / back subcutaneous fat / quality

1 INTRODUCTION

Fat is an essential nutrient in the human diet. However, excessive consumption of fat, and particularly, of certain fatty acids, can cause health risks. Pig meat and meat products are rich sources of fat for human. Two types of fat are especially important in pork and pork products: intramuscular (IMF) and subcutaneous (SCF). Kakovost maščobe avtohtonega krškopoljskega prašiča, rejenega v obogatenem okolju

Raziskave o ugodni sestavi in vsebnosti mišične maščobe (IMF) pri svežem mesu in hkrati podkožnega maščobnega tkiva (SCF) pri predelavi v izdelke znotraj pasme povečujejo zanimanje za avtohtone pasme. Cilj raziskave je bil določiti maščobno-kislinsko sestavo IMF v najdaljši hrbtni mišici (LD) in v pripadajoči SCF pri slovenskih avtohtonih krško-poljskih prašičih. Tip maščobe 42 krško-poljskih prašičev je značilno vplival na 29 od 35 tukaj določenih maščobnih kislin. SCF je vsebovala več nasičenih maščobnih kislin (SFA) in večkrat nenasičenih maščobnih kislin (PUFA) in posledično manj enkrat nenasičenih maščobnih kislin (MUFA) kot IMF. Pri ekstremih je imela 6-10 % IMF v LD večji delež SFA in MUFA in manj PUFA kot 2-3 % IMF. Debelejša SCF (57-67 mm) je vsebovala več SFA in manj PUFA kot tanjša (30-40 mm). Spol in starost sta prav tako vplivala na maščobno-kislinsko sestavo. Maščoba kastratov je vsebovala več MUFA in manj PUFA kot pri svinjkah. Z naraščajočo starostjo je vsebnost MUFA naraščala.

Ključne besede: prašiči / avtohtone pasme / krškopoljski prašič / obogateno okolje / maščobne kisline / mišična maščoba / hrbtna podkožna maščoba / kakovost

The IMF cannot be removed before consumption and both its content and composition have been shown to influence the sensory traits of fresh pork (Ngapo and Gariepy, 2008). In contrast, much of the SCF is removed by trimming, often for use in processed meat products where the processing and storage characteristics of saturated fatty acids (SFA) are desirable (for example, Reichart *et al.*, 2003). The fatty acid composition of pork fat is

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affected by several factors including fat type - location in the carcass, sex (Biederman et al., 2000) and slaughter age (Fischer et al., 2006; De Smet et al., 2004). Decades of pig selection for leanness (Kasprzyk, 2007; Ender, 1994) with the ensuing reduction in IMF content and decreased SCF quality have had a toll on sensory traits (Wood et al., 2003). The search for fat composition and content favourable in IMF for fresh meat quality and at the same time in SCF for processed meat products, both within the same breed, is reflected in the growing interest in native pig breeds (Pugliese and Sirtori, 2012). Native pig breeds are often characterised as having meat of a better eating quality than commercial pigs and fatty carcasses are common (Bonneau and Lebert, 2010; Furman et al., 2010). The aim of this study was to determine the fatty acid profile of the IMF of M. longissimus dorsi (LD) and adjacent SCF from the indigenous Krškopolje (KP) pigs reared in an enriched environment.

2 MATERIALS AND METHODS

Forty two Krškopolje pigs (19 barrows and 23 gilts) were obtained from a local farm at 48.9 ± 7.5 kg (138.5 ± 11.8 days) for fattening under controlled conditions. The pigs were housed in a straw-bedded pen of 150 m² (3.75 m²/animal) with natural ventilation and daylight. Straw bedding was added daily, feed provided once a day (semi-ad libitum 3 kg/animal/day), hay available ad libitum in a profiled rack and fresh water was always available. Feed was wheat, barley and maize-based (14.3 MJ energy/kg DM and 150 g/kg DM crude protein). Hay enrichment provided dietary fibre (287 vs 48 g/kg DM for hay and feed, respectively) and essential α -linolenic acid (43 vs 4 % for hay and feed, respectively). Grain provided linoleic acid (16 vs 54 % for hay and feed, respectively).

Pigs were slaughtered in a commercial slaughterhouse in groups of 9 to 13 animals; the first group after four months in the controlled environment and then four weeks between each following slaughter. The age of animals at slaughter ranged from 236 to 364 days. At 24 h *post mortem*, samples (about 100 g) of lean and adjacent SCF were taken from the LD at the site of the last rib, vacuum packaged and stored at -21 °C. The samples were cut into small pieces (about 1 g), immersed in liquid nitrogen and homogenised (6000 rpm, 10 s; Grindomix 200, Retsch GmbH; Haan, Germany) ensuring the sample temperature did not rise above -2 °C. Homogenised samples were packaged in sealed polyethylene bags and returned to -21 °C until analyses.

The IMF content of the LD was determined by the Weibull-Stoldt method according to the AOAC official method of analysis 991.36 (AOAC International, 1997).

Lipids were extracted from the muscle and adipose tissues by Soxhlet extraction with petroleum ether. Fatty acids were detected as their methyl esters (FAME's; Park and Goins, 1994). Separation of FAME's was performed by gas chromatography using an Agilent 6890 series GC (Agilent Technologies, Wilmington, DE, USA) with an Agilent 7683 Automatic Liquid Sampler, split injector, flame-ionization detector and fused silica capillary column OMEGA-WAX (Supelco, 30 m x 0.32 mm i.d.). Data acquisition and processing was achieved using Agilent GC ChemStation (Rev. A.08.03). Separated FAME's were identified by retention time. Fatty acid composition was calculated using response factors derived from chromatographic standards of known composition (NuChek Prep, Elysian, MN, USA). Results were expressed as percentage of total fatty acids. The atherogenic index (AI) was calculated (Ulbricht and Southgate, 1991).

The statistical model included sex (barrows or gilts) and type of fat (IMF or SCF) as fixed effects and age as covariate. Statistical analysis was carried out using the GLM procedure of the SAS/STAT system (SAS 2008). Within a fat type, statistical differences were determined by t-test.

3 RESULTS

The type of fat had a highly significant effect (P < 0.0001) on the proportions of 29 of the 35 fatty acids measured (major acids are presented in Table 1). A greater proportion of saturated fatty acids (SFA) was found in the SCF than IMF, a consequence of palmitic (C16:0) and stearic (C18:0) acids which were by far the most prevalent of the SFA. The eight other SFA showing significant differences represented less than 4 % of the total fatty acids and therefore had little influence on the total SFA. A greater proportion of PUFA was also found in SCF than in the IMF largely attributed to the n-6 PUFA, and particularly linoleic acid (C18:2n-6). Albeit, less than a tenth the level of n-6 PUFA, the more desirable n-3 PUFA were in greater proportion in the IMF. Consequently, a more desirable n-3/n-6 ratio was found in IMF than SCF, but no differences in PUFA/SFA ratio were observed. In concordance with the lower SFA and PUFA proportions in IMF than SCF, a greater proportion of MUFA were found in the IMF. This difference was largely due to the two most prevalent fatty acids, oleic (C18:1n-9) and palmitoleic (C16:1n-7) acids. The AI was more favourable in the IMF than SCF.

Six fatty acids were different ($P \le 0.05$) between sexes (Table 1). Of these, capric (C10:0) and stearic (C18:0) acids were the only SFA, with the latter having the greater implication in total SFA. Oleic acid (C18:1n-9) was the only MUFA and was in greater proportion in barrows than gilts. At almost 50 %, oleic acid was by far the most abun-

Trait	Fat type		Sex			Age ($b \pm SE$)	P-values		
	IMF	SCF	Barrow	Gilt	RSD	$(\%/day; x 10^{-3})$	Fat type	Sex	Age
Fatty acid (%)									
C14:0	1.28	1.30	1.28	1.29	0.113	0.62 ± 0.42	0.397	0.947	0.136
C16:0	23.44	24.62	24.04	24.02	1.073	-0.93 ± 3.97	<0.001	0.916	0.816
C18:0	10.13	13.66	11.65	12.13	0.913	-10.23 ± 3.38	<0.001	0.028	0.003
C20:0	0.14	0.24	0.19	0.19	0.024	$\textbf{0.20} \pm \textbf{0.09}$	< 0.001	0.470	0.029
SFA	36.96	40.52	38.48	39.00	1.458	-7.38 ± 5.40	<0.001	0.133	0.176
C16:1n-7	4.74	2.54	3.76	3.52	0.548	$\boldsymbol{6.70 \pm 2.02}$	<0.001	0.064	0.002
C18:1n-9	49.22	46.20	48.15	47.27	1.536	6.67 ± 6.67	<0.001	0.017	0.245
C20:1n-9	0.70	1.11	0.92	0.89	0.100	0.07 ± 0.37	<0.001	0.320	0.845
MUFA	55.12	50.59	53.43	52.27	1.557	13.14 ± 5.67	<0.001	0.002	0.025
C18:2n-6c	5.07	7.49	6.08	6.49	0.845	-7.11 ± 3.13	<0.001	0.044	0.026
C20:2n-6	0.14	0.42	0.28	0.28	0.040	-0.52 ± 0.15	<0.001	0.301	<0.001
C20:4n-6	1.48	0.15	0.74	0.89	0.300	$\textbf{2.27} \pm \textbf{1.11}$	<0.001	0.040	0.044
n-6 PUFA	7.28	8.32	7.51	8.09	1.181	-4.83 ± 4.37	<0.001	0.038	0.272
C18:3n-3	0.17	0.38	0.26	0.29	0.036	-0.23 ± 0.13	<0.001	0.006	0.090
C22:5n-3	0.22	0.04	0.11	0.15	0.070	-0.46 ± 0.26	<0.001	0.024	0.078
n-3 PUFA	0.54	0.44	0.46	0.52	0.093	-0.74 ± 0.34	< 0.001	0.003	0.036
PUFA	7.92	8.89	8.08	8.73	1.249	-5.57 ± 4.62	<0.001	0.030	0.232
n-6/n-3	13.81	18.84	16.68	15.98	1.939	8.76 ± 7.18	<0.001	0.131	0.226
PUFA/SFA	0.22	0.22	0.21	0.22	0.037	-0.10 ± 0.14	0.544	0.104	0.487
AI	0.45	0.50	0.48	0.48	0.033	-0.02 ± 0.12	<0.001	0.642	0.862

 Table 1: Effect of sex, fat type and age on fatty acid composition of fat from Krškopolje pigs (LSM)

 Preglednica 1: Vpliv spola, lokacije in starosti na maščobno kislinsko sestavo maščobe pri krškopoljskem prašiču

IMF - intramuscular fat; SCF - subcutaneous fat; RSD - residual standard deviation; b - regression coefficient; SE - standard error

dant of the fatty acids and hence the total MUFA was also greater in barrows than gilts. The gilts had greater proportion of PUFA than barrows, reflecting differences in both n-6 and n-3 PUFA. No differences in the indices of relevance to human nutrition (PUFA/SFA, n-6/n-3 and AI) were observed for sex.

Slaughter age had influence on the fatty acid composition (Table 1). As the slaughter age of the KP pigs increased, so too did the proportion of MUFA, particularly, myristoleic (C14:1n-5) and palmitoleic (C16:1n-7) acids. Neither the total SFA nor the total PUFA were significantly different with age of the KP pigs. However, slaughter age did influence some of the individual unsaturated fatty acids and the proportion of total n-3 PUFA decreased with age. Although the age did not affect the total n-6 PUFA proportion, seven of the individual n-6 PUFA differed with age; three increasing and four decreasing with increasing age. No influence of age was observed on the nutritional indices.

Taking subsets of animals at each extreme of IMF content and SCF thickness, differences in fatty acid con-

tent were observed (Table 2). The KP pigs with high IMF had a higher proportion of SFA and MUFA than the low IMF and a correspondingly lower proportion of PUFA. In fact, all major PUFA were in greater proportion in animals with low IMF. The SCF thickness did not show any effect on MUFA, but animals with thicker SCF had a higher proportion of SFA and lower of PUFA. Superior PUFA/SFA ratios, but less desirable AI were found with lower IMF and with thinner SCF.

4 DISCUSSION

The SCF of Krškopolje pigs had a 3.5 % higher proportion of SFA than the IMF. These findings contrast those of Bosch *et al.* (2012) in Duroc and Franco *et al.* (2006) in Celta pigs who observed higher levels of SFA in IMF than in SCF. In addition, Delgado et al. (2002) found no differences in SFA with type of fat from Hairless Mexican pigs. In the present study, the higher proportion of MUFA, including C18:1 and C16:1 in IMF than in SCF does agree

	IMF (%)		SCF (mm)	P-values		
Trait	2-3	6-10	30-40	57-67	IMF	SCF
Number	7	7	5	5		
IMF (%)	$\textbf{2.7} \pm \textbf{0.3}$	7.3 ± 1.1	4.5 ± 1.5	3.6 ± 1.1	<0.001	0.273
SCF (mm)	48.1 ± 12.5	45.7 ± 5.49	35.1 ± 2.9	61.0 ± 3.4	0.650	<0.001
Weight (kg)ª	142.9 ± 22.1	122.4 ± 2.5	125.7 ± 13.9	154.6 ± 13.7	0.032	0.011
Fatty acid content((%)					
C14:0	1.23 ± 0.12	1.34 ± 0.11	$\textbf{1.24} \pm \textbf{0.09}$	1.39 ± 0.06	0.084	0.017
C16:0	22.66 ± 0.59	24.75 ± 0.86	$\textbf{23.94} \pm \textbf{1.18}$	25.68 ± 0.69	0.002	0.022
C18:0	9.63 ± 0.74	10.55 ± 1.00	12.90 ± 0.64	13.56 ± 0.86	0.076	0.203
C20:0	0.12 ± 0.01	$\boldsymbol{0.14\pm0.01}$	0.23 ± 0.03	0.23 ± 0.02	0.024	0.680
SFA	36.35 ± 0.96	$\textbf{38.07} \pm \textbf{1.59}$	38.97 ± 0.84	41.49 ± 0.89	0.031	0.002
C16:1n-7	5.01 ± 0.89	4.71 ± 0.91	2.69 ± 0.54	2.73 ± 0.46	0.550	0.883
C18:1n-9	47.19 ± 1.48	50.15 ± 1.40	46.96 ± 1.32	46.11 ± 0.45	0.002	0.205
C20:1n-9	0.65 ± 0.05	$\boldsymbol{0.72\pm0.05}$	1.11 ± 0.16	1.14 ± 0.15	0.045	0.739
MUFA	53.34 ± 1.35	55.99 ± 2.15	51.48 ± 1.13	50.62 ± 0.37	0.018	0.143
C18:2n-6	$\boldsymbol{6.47 \pm 0.50}$	4.01 ± 0.91	$\textbf{8.07} \pm \textbf{0.95}$	6.60 ± 0.48	<0.001	0.015
C20:2n-6	0.16 ± 0.03	0.13 ± 0.03	0.44 ± 0.06	0.38 ± 0.03	0.123	0.058
C20:4n-6	$\textbf{2.17} \pm \textbf{0.20}$	$\boldsymbol{0.83 \pm 0.17}$	0.17 ± 0.05	0.14 ± 0.02	<0.001	0.171
n-6 PUFA	$\textbf{9.54} \pm \textbf{0.72}$	5.40 ± 1.08	$\textbf{8.93} \pm \textbf{1.03}$	7.37 ± 0.53	<0.001	0.017
C18:3n-3	0.21 ± 0.04	0.15 ± 0.03	0.41 ± 0.03	$\textbf{0.34} \pm \textbf{0.03}$	0.007	0.008
C22:5n-3	0.26 ± 0.06	0.12 ± 0.03	0.04 ± 0.01	0.04 ± 0.01	0.002	0.955
n-3 PUFA	0.65 ± 0.12	0.42 ± 0.11	$\textbf{0.48} \pm \textbf{0.05}$	$\boldsymbol{0.40\pm0.03}$	0.002	0.020
PUFA	10.31 ± 0.82	5.93 ± 1.16	9.55 ± 1.09	7.89 ± 0.56	<0.001	0.016
n-6/n-3	14.83 ± 1.71	13.03 ± 1.68	18.59 ± 1.34	18.34 ± 0.58	0.071	0.714
PUFA/SFA	$\textbf{0.28} \pm \textbf{0.02}$	0.16 ± 0.03	0.24 ± 0.03	0.19 ± 0.02	<0.001	0.008
AI	0.43 ± 0.02	0.49 ± 0.03	0.48 ± 0.03	$\boldsymbol{0.54\pm0.02}$	0.003	0.007

Table 2: Effect of extremes of IMF content and SCF thickness on fatty acid composition (mean \pm standard deviation)**Preglednica 2:** Vpliv ekstremnih vrednosti za intramuskularno maščobo in podkožno maščobno tkivo na maščobnokislinsko sestavo
(povprečje \pm standardni odklon)

IMF - intramuscular fat; SCF - subcutaneous fat; SD - standard deviation; a Weight - slaughter weight

with findings of Bosch *et al.* (2012). Delgado *et al.* (2002) and Franco *et al.* (2006) also found more C16:1 in IMF than in SCF, but no differences in MUFA content with fat type. As reported by other workers, higher proportions of PUFA, essential linoleic (C18:2n-6) and α -linolenic (C18:3n-3) acids and less arahidonic (C20:4n-6) acid were present in SCF than IMF (Bosch *et al.*, 2012; Franco *et al.*, 2006). While Delgado *et al.* (2002) also found more C18:3n-3 in SCF, no differences were found in PUFA content with fat type.

Sex of the Krškopolje pigs mainly impacted the PUFA content, being higher in gilts than barrows. Salvatori et al. (2008) also reported more unsaturated IMF in gilts than barrows of Casertana pigs. However, both Salvatori et al. (2008) and Pugliese *et al.* (2005; Cinta Senese pigs) found no sex related differences in fatty acid composition of SCF.

Neither Pugliese *et al.* (2005) nor Salvatori et al. (2008) reported any influence of sex on MUFA content.

Slaughter age influenced fifteen of the fatty acids measured here. While daily variations appear relatively small, the age ranged from 236 to 364 days, a difference of more than four months and equivalent to 1.8 % more MUFA in the older animals, including 0.9 % more palmitoleic acid. Decreasing C18:0, C18:2n-6, C20:2n-6 and increasing MUFA with increasing age has also been reported by Bosch *et al.* (2012) in both IMF and SCF. While only one n-3 PUFA differed with age, the total n-3 PUFA and seven of the ten n-6 PUFA varied with age. However, none of the nutritional indices was influenced by age.

Not only was fatty acid composition affected by fat type, but compositional differences were also found within a fat type. The differences in IMF content were in agreement with the findings of Cameron and Enser (1991) that with increasing IMF, the concentrations of SFA and MUFA increase and those of PUFA decrease. In SCF of greater thickness, more C18:0 and less C18:2n-6 and C18:3n-3 were found than in SCF of lower thickness, a trend also observed by Wood *et al.* (1989). Bosch *et al.* (2012) also found similar affects with higher IMF having SFA, MUFA and less PUFA than lower IMF, and thicker SCF having more SFA and less PUFA than thinner SCF.

5 CONCLUSIONS

In conclusion, the type of fat from the Krškopolje pig significantly affected the proportions of 29 of the 35 fatty acids measured here. The SCF contained higher proportions of SFA and PUFA and correspondingly less MUFA than the IMF. Taking the extremes, the IMF at 6–10 % of the LD had higher proportions of SFA and MUFA and less PUFA than the IMF at 2–3 %. The thicker SCF (57– 67 mm) was higher in SFA and lower in PUFA than thinner SCF (30–40 mm). Sex and age also effected on fatty acid composition. In particular, the fat from barrows was higher in MUFA and lower in PUFA than that from gilts, and with increasing age, MUFA content increased.

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