

## Performance and fatty acid profile in the selected tissues of Polish Large White breed and crossbred pigs fed a diet enriched in omega-3 fatty acids\*

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The effect of breed on fatty acid (FA) content of the selected tissues of the pigs fed a diet enriched in omega-3 FA was determined in sixteen Polish Large White (PLW, n=8) and ♂Duroc x ♀Polish Large White x ♂Danish Landrace (n=8) pigs. From 70 to 110 kg body weight (BW) the animals were fed a diet in which 9% of ME originated from energy of oil mixtures: 2.0% linseed, 0.5% rapeseed, 0.5% fish, which introduced to the diet C18:3 n-3 (ALA), C20:5 n-3 (EPA), C22:5 n-3 (DPA) and C22:6 n-3 (DHA) acids. The pigs were slaughtered at approximately 110 kg BW. The *Musculus longissimus dorsi* (MLD) and subcutaneous fat (ST) samples were collected for analysis of extract ether (%) and FA (g per 100 g of tissue) content. The analyzed breeds had a similar performance and backfat thickness, but they differed in the intramuscular fat content. MLD of crossbred pigs had higher content of SFA, MUFA and PUFA (including ALA, EPA, DPA and DHA) than PLW pigs, whereas subcutaneous fat of crossbred pigs had lower content of MUFA (including C16:1 n-7 and C18:1 n-9) and higher content of PUFA (including n-3 PUFA) than PLW pigs. Intramuscular fat of both breeds had similar ratio of PUFA/SFA (average 0.44), whereas the ratio of n-6 PUFA/n-3 PUFA ( $\Sigma n-6/\Sigma n-3$ ) was lower (more favorable) in crossbred than PLW pigs (3.73 vs. 5.80). This was caused by the higher n-3 PUFA content in MLD, which was related to the higher intramuscular fat content in crossbred pigs. Moreover, crossbred pigs had more favorable ratio of PUFA/SFA in subcutaneous fat than PLW pigs (0.58 vs. 0.50), whereas the ratio of  $\Sigma n-6/\Sigma n-3$  was similar in both pig breeds (average 2.83).

**KEY WORDS:** omega-3 fatty acids / pigs / subcutaneous fat / intramuscular fat / *Musculus longissimus dorsi*

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Meat characteristics considered to be of paramount importance for consumers include intramuscular fat (IMF) content, since it affects sensory and eating quality of pork, and the fatty acid (FA) profile, determining its health-promoting value. Selection of animals towards decreased backfat thickness has resulted in the reduction of total fat content in the carcass, including intramuscular fat [13]. For this reason IMF content in muscles (e.g. *longissimus dorsi*) of fatteners characterised by high carcass meatiness is 0.5-1.5% [9]. In contrast, it is assumed that optimal IMF content in muscles should be 2-3% [11]. Fat content and the fatty acid profile in porcine tissues/meat are influenced e.g. by fatty acid contents in feed [5, 18] and by the pig breed [12, 15]. Results presented by Burkett et al. [2] and Raj et al. [15] indicate that pigs exhibiting a greater overall carcass fatness have higher levels of intramuscular fat, which contains more saturated fatty acids (SFA) and less polyunsaturated fatty acids (PUFA) in comparison to that of leaner pigs. Other researchers reported that both lean and fat pigs similarly respond to an addition of fats to the administered feed, altering the fatty acid profile and health-promoting properties of pork [14].

Dependencies between subcutaneous fat, intramuscular fat content and the FA profile in tissues of pigs fed a standard diet have been thoroughly described. In contrast, scarce studies aimed at the determination of these dependencies in tissues of pigs of different breeds/genotypes fed a diet supplemented with omega-3 fatty acids.

The aim of this study was to determine the effect of breed in pigs differing in intramuscular fat contents, but having comparable overall carcass fatness, fed a diet enriched with C18:3 *n*-3 (ALA), C20:5 *n*-3 (EPA), C22:5 *n*-3 (DPA) and C22:6 *n*-3 (DHA) fatty acids on their content (g/100 g) in the *longissimus dorsi* muscle and subcutaneous fat.

## Material and methods

The experiment was conducted on 16 gilts of the Polish Large White breed (PLW,  $n=8$ ) and crosses ♂Duroc x ♀(Polish Large White x Danish Landrace) ( $n=8$ ), which had comparable carcass fatness levels, but differed in terms of intramuscular fat. Pigs from 25 to 110 kg body weight were housed on a slated floor in individual pens of 2.6 m<sup>2</sup> equipped with nipple drinkers. All pigs incorporated into the study were free from genes predisposing to meat defects. Gilts within breed/genotype were sired by one boar and came from related dams (half-sibs). Animal handling in the course of the study followed the requirements of legal standards determining the conditions and methods of experimentation on animals.

From 25 to 70 kg body weight animals were fed standard diet (13.2 MJ ME and 8.2 g digestible lysine), next up to 110 kg BW they received pelleted experimental feed composed of barley (36.5%), maize (36%), wheat (10%), soybean meal (8%) and rapeseed meal (4%) and enriched with a mixture of minerals and vitamins (2.5%) supplementing 1 kg feed with 2.8 g Ca, 0.07 g P, 60 mg Fe, 50 mg Zn, 30 mg Cu, 30 mg Mn, 0.30 mg J, 0.20 mg Se, 1500 IU vitamin A, 300 IU vitamin D<sub>3</sub>, 150 mg vitamin E, 2.0 mg vitamin K<sub>3</sub>, 2.0 mg vitamin B<sub>1</sub>, 2.5 mg vitamin B<sub>2</sub>, 2.0 mg vitamin B<sub>6</sub>, 0.02 mg vitamin B<sub>12</sub>, 0.11 mg biotin, 0.6 mg folic acid, 15 mg nicotinic acid, 10 mg D-calcium pantothenate, 500 mg choline chloride, 2.63 g lysine, 0.68 g methionine and 0.98 g threonine. In the experimental feed

9% metabolisable energy came from a mixture of linseed (2%), rapeseed (0.5%) and fish oils (0.5%), supplying acids: C18:3 *n*-3 (ALA), C20:5 *n*-3 (EPA), C22:5 *n*-3 (DPA) and C22:6 *n*-3 (DHA). The fatty acid composition of feed is given in Table 1.

**Table 1**  
Fatty acid (FA) content in the diet (%)

Fatty acids	%
ΣSFA	16.61
ΣMUFA	29.54
ΣPUFA	52.15
ΣPUFA/ΣSFA	3.14
C16:0	12.36
C16:1 <i>n</i> -7	0.92
C18:0	2.83
C18:1 <i>n</i> -9	24.0
C18:2 <i>n</i> -6	35.0
C18:3 <i>n</i> -3	15.2
C20:4 <i>n</i> -6	0.46
C20:5 <i>n</i> -3	0.69
C22:5 <i>n</i> -3	0.09
C22:6 <i>n</i> -3	0.96
Σ <i>n</i> -6 FA	35.13
Σ <i>n</i> -3 FA	16.98
18:2 <i>n</i> -6/18:3 <i>n</i> -3	2.30
Σ <i>n</i> -6/Σ <i>n</i> -3	2.07

In order to protect the feed against oxidation of long-chain polyunsaturated fatty acids vitamin E was added as an antioxidant.

Digestibility of chemical components in the experimental feed was tested on 4 gilts from each breed group at 80-90 kg BW using the reference method with chromium oxide green according to Fenton and Fenton [7]. Metabolisable energy of feed was estimated based on digestible nutrients [16].

When they reached approx. 110 kg body weight the pigs were slaughtered after electric stunning at an abattoir of the Institute of Animal Physiology and Nutrition PAS. Half-carasses were cooled at 4°C for 24 h. Backfat thickness was measured on the right half-carass at 5 points following the SKURTCh methodology, next it was weighed and dissected. The longissimus dorsi muscle (*Musculus longissimus dorsi* – MLD), subcutaneous fat and meat in the half-carass were dissected and weighed. Carcass meatiness was determined as the ratio of meat in the half-carass to its cold weight and next the result was expressed in percent. The carcass was divided into edible/soft parts (meat and fat) and inedible parts (skin and bones). MLD and subcutaneous fat were ground separately and a 500g sample was homogenised, placed in a plastic bag and stored at –20°C until analysis of the chemical composition of tissues and the fatty acid profile. Edible/soft tissues from the carcass were ground, while inedible tissues after weighing were autoclaved for 8 h at 130°C at a pressure of 1.2 atm. After these tissues were ground and homogenised samples were collected for analyses of the chemical composition of the carcass.

Fat samples from feed, the longissimus dorsi muscle and subcutaneous fat were extracted with a solution of chloroform and methanol (2:1) according to Folch et al. [8]. The fatty acid profile of methyl esters was assayed using a Shimadzu GC-2010AF gas chromatograph with a flame ionization detector (FID) equipped with a 60m capillary column (BPX70) of 0.25 mm inner diameter and 0.25 µm coating thickness. Helium was the carrier gas. The applied split ratio was 1:100. The temperature in the detector and injector was 260°C. Initial temperature of the column was 140°C, it was maintained for 1 min, afterwards it increased to 200°C at a rate of 4°C/min, followed by a further increase to 220°C at a rate of 1°C/min. The entire process lasted for 36 min. Individual peaks of fatty acids were identified by comparison to the Supelco 37 Component FAME Mix reference standard. Based on the results quantitative and qualitative analyses of fatty acids were performed.

Statistical analysis was performed using Statgraphics Centurion XVI software (version 16.1.18, 2011). Differences between pig breeds in terms of the investigated traits were assessed using one-way analysis of variance ANOVA.

## Results and discussion

The experimental feed contained (in 1 kg) 13.4 MJ ME, 146 g digestible protein and 7.4 g dietary standardised ileal digestible lysine, while the proportion of lysine to metabolisable energy was 0.55 g/MJ ME. Pigs of both groups consumed comparable amounts of feed, had similar daily weight gains and feed conversion ratios (2.92 kg, 1170 g and 2.47 kg/kg, respectively) – Table 2. Results of this study are consistent with those reported e.g. by Eckert et al. [6], Skiba et al. [17] and Wojtasik et al. [20]. In turn, Michalska

**Table 2**

Performance of pig and carcass characteristic in the experimental period (70-110 kg BW)

Indices	Breed		SE	Significance
	PLW	crossbred <sup>1</sup>		
Body weight (kg)	112.5	111.4	2.932	ns
Feed intake (kg/day)	2.92	2.92	0.060	ns
ADG <sup>2</sup> (g)	1197	1143	20.877	ns
FCR <sup>3</sup> (kg)	2.45	2.49	0.094	ns
Cold carcass weight (kg)	88.7	84.7	2.458	ns
Average backfat thickness (mm)	23.12	23.77	1.127	ns
Meat content in carcass (%)	60.18	60.42	0.546	ns

<sup>1</sup>Crossbred pigs: Duroc x (Polish Large White x Danish Landrace); <sup>2</sup>ADG – average daily gain; <sup>3</sup>FCR – feed conversion ratio; ns – not significant

[10] reported a better feed conversion ratio per 1 kg body weight gain in F1 crosses than in purebred pigs, which was caused by the heterosis effect.

The PLW pigs and crossbreds had similar carcass weights, backfat thickness and carcass leanness (mean 86.7 kg, 23.45 mm and 60.30%, respectively). Pigs from both groups were characterised by comparable contents of fat, protein and ash in the carcass (mean 220, 174 and 30 g/kg; Table 3). Similar results were presented by Czarniecka-Skubina et al. [3] and Blicharski et al. [1].

**Table 3**

Cold carcass weight (kg) and chemical composition of carcass (g/kg of carcass)

Indices	Breed		SE	Significance
	PLW	crossbred <sup>1</sup>		
Cold carcass weight	88.7	84.7	2.46	ns
Protein content	173	174	2.71	ns
Ether extract content	221	219	5.39	ns
Ash content	28	31	0.80	ns
Water content	568	569	5.30	ns

<sup>1</sup>Crossbred pigs: Duroc x (Polish Large White x Danish Landrace); ns – not significant

Animals from both analysed groups had longissimus dorsi muscles of similar weight (mean 2603 g, Table 4), but PLW pigs were characterised by a lower ( $P \leq 0.01$ ) intramuscular fat content in MLD than crossbred gilts (mean 0.79 vs. 1.83%). The results are consistent with those reported in studies conducted by Doran et al. [4] and Czarniecka-Skubina et al. [3].

Greater intramuscular fatness in MLD of crossbred pigs resulted in increased FA contents in comparison to PLW pigs. The longissimus dorsi muscle in crossbred pigs contained a two-fold greater amounts of saturated fatty acids (SFA,  $P \leq 0.01$ , including C16:0 and C18:0,  $P \leq 0.001$ ) and monounsaturated fatty acids (MUFA, including C16:1 *n-7* and C18:1 *n-9*,  $P \leq 0.01$ ) as well as a three-fold greater content of polyunsaturated fatty acids (PUFA, including linoleic acid – LA, C18:2 *n-6* and  $\alpha$ -linolenic acid – ALA, C18:3 *n-3*,  $P \leq 0.001$ ) in comparison to PLW pigs. Moreover, in pigs exhibiting greater MLD fatness (crossbreds) contents of arachidonic acid (AA, C20:4 *n-6*,  $P \leq 0.01$ ) and eicosapentaenoic acid (EPA, C20:5 *n-3*,  $P \leq 0.001$ ) were two-fold greater, that of docosapentaenoic acid (DPA, C22:5 *n-3*) four-fold greater ( $P \leq 0.001$ ), while that of docosahexaenoic acid (DHA, C22:6 *n-3*) six-fold greater ( $P \leq 0.001$ ) in comparison to gilts with low fat contents in MLD (PLW). Presented results are consistent with the studies reported by Skiba et al. [18], who showed a positive dependence between contents of polyunsaturated fatty acids from the *n-3* family and fat content in the longissimus dorsi muscles in pigs.

**Table 4**

Weight of the *longissimus dorsi* muscle (MLD, g), content of intramuscular fat (IMF, %) and total content (g/100 g of tissue) of fatty acids (FA), SFA, MUFA, PUFA and particular FAs in the MLD

Indices	Breed		SE	Significance
	PLW	crossbred <sup>1</sup>		
MLD	2638	2568	107.82	ns
IMF	0.79	1.83	0.169	xx
ΣFA	0.702	1.643	0.152	xxx
ΣSFA	0.270	0.614	0.060	xx
ΣMUFA	0.322	0.731	0.072	xx
ΣPUFA	0.104	0.292	0.025	xxx
ΣPUFA/ΣSFA	0.40	0.48	0.032	ns
C16:0	0.160	0.375	0.036	xxx
C16:1 <i>n</i> -7	0.020	0.039	0.005	xx
C18:0	0.089	0.220	0.022	xxx
C18:1 <i>n</i> -9	0.267	0.600	0.060	xx
C18:2 <i>n</i> -6	0.070	0.194	0.016	xxx
C18:3 <i>n</i> -3	0.012	0.039	0.004	xxx
C20:4 <i>n</i> -6	0.012	0.025	0.003	xx
C20:5 <i>n</i> -3	0.003	0.007	0.001	xxx
C22:5 <i>n</i> -3	0.002	0.009	0.001	xxx
C22:6 <i>n</i> -3	0.001	0.006	0.001	xxx
Σ <i>n</i> -6 FA	0.088	0.226	0.020	xxx
Σ <i>n</i> -3 FA	0.018	0.060	0.005	xxx
18:2 <i>n</i> -6/18:3 <i>n</i> -3	5.96	5.04	0.251	x
Σ <i>n</i> -6/Σ <i>n</i> -3	5.80	3.73	0.396	xx

<sup>1</sup>Crossbred pigs: Duroc x (Polish Large White x Danish Landrace); ns – not significant; xxx – significant at  $P \leq 0.001$ ; xx – significant at  $P \leq 0.01$ ; x – significant at  $P \leq 0.05$

In contrast to crossbred gilts, MLD in PLW gilts had a higher ratio of respective acids, i.e. a value inferior to that recommended by WHO [19], as it was 18:2 *n*-6/18:3 *n*-3 ( $P \leq 0.05$ ) and Σ*n*-6/Σ*n*-3 ( $P \leq 0.01$ ) (5.96 and 5.80 vs. 5.04 and 3.73). This was caused by a lower IMF content, resulting in lower amounts of *n*-3 PUFA in PLW pigs. In turn, the PUFA/SFA ratio was comparable in both analysed pig groups (mean 0.44).

These results along with the findings reported by other authors indicate that not only fed composition [5, 18], but also pig breed [12, 15] influence the qualitative and quantitative composition of fatty acids accumulated in the carcass.

Both analysed pig groups were characterised by comparable weights of subcutaneous fat and fat contents (ether extract) in subcutaneous fat (mean 5.89 kg and 75.07%, respectively; Table 5). Similar results were presented by Doran et al. [4] and Czarniecka-Skubina et al. [3].

Subcutaneous fat in the Large White Polish gilts contained greater ( $P \leq 0.01$ ) amounts of MUFA (including approx. 25% more ( $P \leq 0.01$ ) C16:1 *n*-7 and approx. 6% more ( $P \leq 0.05$ ) C18:1 *n*-9) and arachidonic acid in comparison to crossbred gilts. In turn, in subcutaneous fat of crossbred gilts greater ( $P \leq 0.05$ ) contents of PUFA were recorded (including ALA,

**Table 5**

Weight of the subcutaneous fat (ST, kg), fat content (%) and total content (g/100 g of tissue) of fatty acids (FA), SFA, MUFA, PUFA and particular FAs in the ST

Indices	Breed		SE	Significance
	PLW	crossbred <sup>1</sup>		
ST	6.07	5.70	0.239	ns
Fat	75.28	74.85	0.729	ns
ΣFA	67.755	67.367	0.657	ns
ΣSFA	24.752	24.694	0.525	ns
ΣMUFA	30.316	28.202	0.399	xx
ΣPUFA	12.300	14.276	0.481	x
ΣPUFA/ΣSFA	0.50	0.58	0.026	x
C16:0	14.962	14.395	0.305	ns
C16:1 <i>n</i> -7	1.428	1.136	0.063	xx
C18:0	8.607	9.222	0.280	ns
C18:1 <i>n</i> -9	26.129	24.702	0.363	x
C18:2 <i>n</i> -6	8.100	8.575	0.965	ns
C18:3 <i>n</i> -3	2.879	3.187	0.074	x
C20:4 <i>n</i> -6	0.323	0.107	0.052	x
C20:5 <i>n</i> -3	0.063	0.073	0.003	x
C22:5 <i>n</i> -3	0.162	0.183	0.007	x
C22:6 <i>n</i> -3	0.100	0.120	0.004	xx
Σ <i>n</i> -6 FA	8.917	10.244	0.433	x
Σ <i>n</i> -3 FA	3.204	3.563	0.082	xx
18:2 <i>n</i> -6/18:3 <i>n</i> -3	2.82	3.03	0.124	ns
Σ <i>n</i> -6/Σ <i>n</i> -3	2.79	2.87	0.120	ns

<sup>1</sup>Crossbred pigs: Duroc x (Polish Large White x Danish Landrace); ns – not significant; xx – significant at  $P \leq 0.01$ ; x – significant at  $P \leq 0.05$

EPA and DPA,  $P \leq 0.05$  and DHA,  $P \leq 0.01$ ) in comparison to subcutaneous fat of PLW gilts. Similarly, in a study by Burkett et al. [2], subcutaneous fat of pigs with greater intramuscular fat contents had lower levels of MUFA and higher levels of SFA. This difference resulted from the administered diet, which in turn affected the de novo synthesis pathway. In our experiment we used feed enriched with a mixture of oils as a source of *n*-3 PUFA, while in the study of Burkett et al. [2] it was supplemented with lard, which is a rich source of saturated fatty acids. This indicates a significant effect of feed composition on the profile and contents of fatty acids in porcine tissues.

In subcutaneous fat of crossbred pigs a greater ( $P \leq 0.05$ ) proportion of PUFA/SFA was found in comparison to that of PLW pigs (0.58 vs. 0.50), whereas the ratios of 18:2 *n*-6/18:3 *n*-3 and Σ*n*-6/Σ*n*-3 were comparable in both breeds.

Summing up it was stated that an improvement of the quantitative and qualitative composition of fatty acids in porcine meat and fat, thanks to the supplementation of feed with *n*-3 PUFA, is easier to attain in pigs with a greater intramuscular fat content.

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