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The influence of dietary source of fatty acids on chemical composition of the body and utilization of linoleic and linolenic acids by pigs*

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The study was carried out on 36 gilts (♀Polish Large White x ♂Danish Landrace) grown from 60 to 105 kg body weight (BW). Experimental diets were composed on the basis of the control diet (C), replacing 13% of its energy by energy from linseed oil (group/diet L), rapeseed oil (group/diet R), beef tallow (group/diet T) or fish oil (group/diet F). All diets contained similar amount of metabolizable energy and apparent ileal digestible lysine, but had different ratio of PUFA to SFA and of C18:2*n*-6 to C18:3*n*-3. Pigs were slaughtered at 105 kg BW. Protein, fat and fatty acids (FA) content of the pigs' empty body was determined by slaughter method. Final empty body of pigs (at 103.6 kg) of individual groups contained similar amount of protein, fat and total FA, but different content of individual FA. Increasing the daily consumption of C18:2*n*-6 and C18:3*n*-3 acids enhanced their deposition in the body (from 16.18 to 27.70 and from 1.16 to 33.82 g/day, respectively). Linear correlations were identified between the intake of linoleic and linolenic acids and their deposition in the empty body (r = 0.96 and r = 0.99, respectively). Efficiency of utilization of C18:2*n*-6 was lower than that of C18:3*n*-3 (coefficient 0.67 *vs.* 0.79, respectively).

KEY WORDS: body / fat / linoleic acid / linolenic acid / pigs / protein

Breed, sex, age, body mass, environmental temperature, rearing system, feeding system and diet composition all affect the fat and fatty acids (FA) content of pig tissues.

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Although the literature delivers abundant information on the relationship between pigs feeding and FA profile in their individual tissues (e.g. in backfat or *longissimus dorsi* muscle, these results can not be compared to other tissues [Barowicz *et al.* 2002, Więcek *et al*. 2004, Pascual *et al.* 2007, Flachowsky *et al.* 2008, Wood *et al.* 2008].

The classic model of growth lets to estimate the growth rate precisely, as well as protein and fat deposition in pigs body on the basis of intake of available energy and individual nutrients [Whittemore and Fawcett 1976, de Lange 1995]. However, the relationship between FAs intake and their deposition in tissues and in the whole body is still not established definitely. Lizardo *et al*. [2002] as first attempted at compiling the mathematical relationship between feeding and deposition of the most important FAs in subcutaneous and perinephric fat. They, however, concluded that compiling the model without full information regarding deposition of FA in the whole body is of low precision and requires further investigations.

In this study an attempt was made at estimating the relationship between the intake of linoleic and linolenic FAs and their deposition in the whole body. Both are of essential importance in many metabolic processes, but cannot be synthesized by pigs. That is why they are considered exogenous and must be supplied in the diet.

The aim of the present investigation was to separate from energy conversions in the body a part which animal deposits as fat and fatty acids. Furthermore, an attempt was made at determining the intake of linoleic and linolenic acids from the diet on their utilization efficiency by pigs. Information received will be used to study the nutritive model of FAs in growing pigs. Moreover, the results will correct the data on nutritive requirements of pigs and improve the quality and health-promoting properties of pork.

It was assumed that replacing part of the dietary metabolizable energy by fats with various content of fatty acids, will not change the amount of fat and total FA in the whole body, but FA composition of the body fat will be altered.

Material and methods

The experiment was carried out on 36 crossbred gilts (♀Polish Large White × ♂Danish Landrace) of a body weight (BW) from 60 to 105 kg. Origin of piglets minimised genetic variability as they were the offspring of one Danish Landrace boar and five half-sibling Large White sows. One piglet from each litter was allocated to each experimental group. Pigs were kept in individual pens (2.6 m^2) on concrete floor, without straw, with free access to drinking water, in thermo-neutral air temperature of 18-20°C and humidity of 60-70%. Animals of control group (diet/group C) were fed diet containing barley, wheat, maize, post-extractive soyabean and post-extractive rapeseed oil meal, supplemented with mineral-vitamin mixture enriched with crystalline amino acids. In the experimental diets, approximately 13% of ME energy of diet C was replaced with energy of linseed oil (diet/group L), rapeseed oil (diet/group R), beef tallow (diet/group T) or fish oil (diet/group F). All diets were

Table 1. Ingredients, chemical composition and nutritive value of diets

Item	Diet/group[1]				
	C	L	R	T	F
Ingredient (g·kg^{-1})					
barley	305	325	325	325	325
wheat	300	360	360	360	360
maize	250	-	-	-	-
soyabean meal	40	40	40	40	40
rapeseed meal	80	80	80	80	80
wheat bran	-	120	120	120	120
linseed oil	-	50	-	-	-
rapeseed oil	-	-	50	-	-
beef tallow	-	-	-	50	-
fish oil (cod liver oil)	-	-	-	-	50
Premix A[2]	25	-	-	-	-
Premix B[2]	-	25	25	25	25
Chemical composition (g·kg^{-1})					
dry matter	891	903	902	900	890
ash	38	41	41	44	45
organic matter	853	862	857	856	845
crude protein	158	162	162	163	160
ether extract	24.8	67.4	67.7	62.7	78.0
crude fibre	34.0	40.2	42.9	42.2	43.2
nitrogen-free-extractives	674	592	584	588	564
Content (g·kg-1)					
digestible crude protein	134	135	136	138	134
apparent ileal digestible[3]					
lysine	8.17	8.19	8.23	8.20	8.23
methionine	2.83	2.81	2.82	2.80	2.82
threonine	5.59	5.49	5.50	5.55	5.50
thryptophan	1.46	1.45	1.43	1.47	1.42
P, digestible	1.68	1.65	1.56	1.58	1.62
Ca	4.58	4.58	4.53	4.55	4.61
Na	1.03	1.03	1.00	1.10	1.05
metabolizable energy (MJ·kg^{-1})[4]	13.30	13.58	13.56	13.58	13.60
Lys:ME (g MJ^{-1})	0.61	0.60	0.61	0.60	0.61

[1]C – control group; L – linseed oil; R – rapeseed oil; T – beef tallow; F – fish oil (cod liver oil).
[2]Premix A and B as 2.5% of the diet provided the required amount of minerals, essential amino acids and vitamins.
[3]Calculated according to CVB [1995].
[4]Calculated on the basis of digestible nutrient components [FBN, 2003].

iso-energetic (13.52 MJ ME kg^{-1}) and iso-protein (135g kg^{-1} digestible protein). They were balanced in relation to the content of ileal digestible amino acids, but differed in ratios of PUFA:SFA and linoleic:linolenic acids (Tab. 1 and 2).

Animals were fed restrictively (85% of *ad libitum*) twice a day. Feed allowance was changed weekly regarding the BW of pigs. At 60 kg BW, six pigs were slaughtered (animals "0") and protein, fat and fatty acids contents of their body were determined.

Remaining 30 pigs were allotted into five groups (six animals in each) and fed C, L, R, T or F diet.

The apparent digestibility of energy and chemical ingredients of feeds was determined after three-day faeces collection in all pigs weighing about 80 kg, with chromium oxide as a marker. Metabolizable energy of diets was calculated based on digestible nutrient components, taking into account correction for the content of digestible protein, fat, starch and simple sugars of the organic matter [FBN, 2003].

The pigs were slaughtered at 105 kg BW and their bodies were analysed for protein, fat and fatty acids. After slaughter, the carcass and non-carcass parts (viscera, blood and hair) were weighed separately. Carcasses were chilled overnight. Next, the right carcass-sides were partially dissected into soft tissues (meat and fat) and bones. Non-edible parts (head, legs, skin and bones) and viscera with collected blood were autoclaved for 12 h. Further, soft tissue and non-edible parts were homogenized and the samples were taken to determine protein, fat and fatty acids. Retention of protein, fat and fatty acids in the body during growth from 60 to 105 kg was estimated using comparative slaughter method [Kotarbińska 1971]. The gain of these body components over the experimental period was calculated as the difference between the determined final values and the estimated initial values of the empty body.

Chemical composition of diets and protein and fat contents of the animals' body were determined using standard methods [AOAC 1995]. Chromium oxide in the diets and faeces was determined according to Fenton and Fenton [1979]. The FA profile was established in homogenized samples with methyl esters method [Folch *et al.* 1957]. Fatty acids methyl esters were separated by gas chromatography on GC-2010 AF SCHIMADZU gas chromatograph, equipped with a BPX70 capillary column (60 m x 0.25 mm × 0.25 μm) with helium as a carrier gas. The total content of fatty acids was calculated as 90% of ether extract [Kratz 2003]. The intake of digestible FAs from a diet was calculated regarding their intestinal digestibility [Jřrgensen *et al*. 1992, 1993]. The protein, fat and fatty acids contents of the empty body weight (EBW) were calculated as a sum of their content of soft tissues, non-edible parts and viscera with blood. Efficiency of utilization of C18:2*n*-6 (LA) and C18:3*n*-3 (ALA) were calculated as: daily deposition (g) of LA or ALA in the EBW/daily intake (g) of LA or ALA.

Fatty acids source of diets was used as the main factors in the variance analysis, and the Tuckey-test was used to compare differences between means. All calculations were performed using STATGAFICS Centurion Software version XV [2005].

Results and discussion

The mean EBW of "0" animals amounted 58.8±1.26kg and contained: protein 9.41±0.34 kg, ether extract 6.87±0.75 kg, FAs 6.18±0.69 kg, LA 861±34.44g and ALA 117±5.85g ALA.

Fatty acids profile, ratios of PUFA:SFA and of linoleic:linolenic acid content of diets are presented in Table 2.

Table 2. Fatty acids profile of diets (% total FA)

Item	Diet/group[1]				
	C	L	R	T	F
SFA	24.65	17.01	11.92	43.33	19.59
MUFA	28.61	22.00	49.46	30.64	31.43
PUFA	43.99	58.69	37.00	22.86	40.69
18:2n-6 (ALA)	40.56	27.70	28.39	19.60	19.61
18:3n-3 (LA)	2.76	30.53	7.55	3.06	8.88
PUFA/SFA	1.78	3.45	3.10	0.53	2.08
18:2n-6/18:3n-3	14.70	0.91	3.76	6.41	2.21

[1]C – control group; L – linseed oil; R – rapeseed oil; T – beef tallow; F – fish oil (cod liver oil).

Table 3. Growth performance of pigs (60-105 kg body weight)

Item	Diet/group[1]					Mean	SE
	C	L	R	T	F		
Feed intake (kg·day^{-1})	2.5	2.5	2.5	2.5	2.5	2.5	0.000
ME (MJ·day^{-1})	33.4	33.9	33. 5	33.9	34.0	33.85	0.236
Intake digestible lysine (g·day^{-1})	20.4	20.5	20.6	20.5	20.6	20.51	0.058
Average daily gain (g)	930	985	943	969	973	961	21.520
Feed conversion ratio (kg·kg^{-1})	2.69	2.54	2.65	2.58	2.57	2.61	0.030

[1]C – control group; L – linseed oil; R – rapeseed oil; T – beef tallow; F – fish oil (cod liver oil)
SE – standard error.

The fattening results of pigs were not significantly influenced by experimental treatment - fat sources (Tab. 3). In the present work, the influence of diets with various fatty acids content on daily gain and feed conversion ratio was not affirmed and corroborates the earlier studies by Nguyen *et al*. [2003] and Mitchaothai *et al.* [2007] in which the experimental pigs were fed either restricted amount of feed or fed *ad lib.*

As assumed, replacing part of energy of the control diet by fatty acids of different sources did not affect protein, fat and total fatty acids content of the body of pigs, confirming that experimental diets were properly balanced concerning their nutritive value and energy content [CVB 1995]. The EBW of pigs slaughtered at 105 kg BW contained similar amount of protein, ether extract and total FAs, but differed ($P<0.01$) in content of linoleic and linolenic acid (Tab. 4). Diet L had produced the highest amount of linolenic acid and diet R and L the highest amount of linoleic acid.

Increasing daily intake of LA and ALA led to their increased deposition in the body (Tab. 5). The efficiency of utilization of LA was smaller than of ALA (mean 0.67 *vs.* 0.79, respectively) and depended not only on their intake but also on their source.

In the present study the efficiency of utilization of linolenic acid was higher than that of linoleic, differing from results showed by Kloarega *et al.* [2005, 2007]

Table 4. The protein, ether extract, total FAs, linoleic and linolenic acid content in the empty body weight (EBW) pigs slaughtered at 105 kg body weight

Item	Diet/group[1]					Mean	SE
	C	L	R	T	F		
EBW	103	104	104	103	103	103.5	2.21
Protein (kg)	16.91	17.39	16.96	16.87	16.94	17.01	0.35
Ether extract (kg)	18.12	19.06	18.74	18.47	18.75	18.63	0.65
Total FAs (kg)	16.29	17.15	16.87	16.62	16.88	16.76	0.59
Linoleic acid (g)	1754^{C}	2162^{A}	2246^{A}	1670^{C}	1910^{B}	1948	30
Linolenic acid (g)	175^{D}	1808^{A}	555^{B}	292^{C}	618^{B}	690	39

[1]C – control group; L – linseed oil; R – rapeseed oil; T – beef tallow; F – fish oil (cod liver oil).
SE – standard error.
[AB...]Within rows means bearing different superscripts differ significantly at P<0.01.

Table 5. The linoleic and linolenic acid balance (g·day^{-1}) in empty body of pigs (60-105 kg body weight)

Fatty acid	Diet/group[1]					Mean	SE
	C (n=6)	L (n=6)	R (n=6)	T (n=6)	F (n=6)		
Linoleic acid (18:2*n*- 6)							
intake	21.66^{D}	40.10^{A}	41.208^{A}	26.17^{C}	32.56^{B}	32.34	0.602
deposition	17.86^{C}	26.02^{A}	27.70^{A}	16.18^{C}	21.95^{B}	21.74	1.061
efficiency of utilization[2]	0.825^{A}	0.673^{B}	0.671^{B}	0.619^{C}	0.674^{B}	0.672	0.039
Linolenic acid (18:3*n*-3)							
intake	1.44^{E}	43.28^{A}	10.72^{C}	4.00^{D}	$14.44^{\B}$	14.78	0.380
deposition	1.16^{D}	33.82^{A}	8.76^{B}	3.50^{C}	10.02^{B}	11.90	0.700
efficiency of utilization[2]	0.810^{B}	0.781^{C}	0.817^{B}	0.875^{A}	0.694^{D}	0.792	0.047

[1]C – control group; L – linseed oil; R – rapeseed oil; T – beef tallow; F – fish oil (cod liver oil).
[2]Efficiency of utilization were calculated as: daily deposition (g) in the empty body/daily intake (g).
SE – standard error.
[AB...]Within rows means bearing different superscripts differ significantly at P<0.01.

who based on comparative slaughter method and found that efficiency of utilization of ALA was lower than of LA. The difference could result from differences in the rearing and feeding system used. In the present experiment the animals were kept under thermo-neutral conditions and fed restrictively a diet with various amount of linoleic and linolenic acid. However, Kloarega *et al.* [2005] used a diet fed according to various feeding systems (100, 80 and 70% of *ad libitum*). Moreover, environmental temperature in their study was much higher.

Relationship between above factors expressed as Pearson correlation amounted at $r = 0.96$ for LA and $r = 0.99$ for ALA (not tabulated). A high correlation was identified between the daily intake and LA and ALA content of the body of pigs, being in accordance with Nguyen *et al*. [2003] who, however, unlike as in the present study, analysed a correlation between amount of exogenous fatty acids in the diet (g·MJ metabolizable energy) and their content (% of total FA) in the subcutaneous and intramuscular fat

only. However, when Nguyen *et al*. [2003] calculated these relationships based on literature data, estimated correlation occurred much lower. Discrepancy resulted probably from increased variation due to differences in experimental design and conditions which led to a decrease in the correlation coefficients.

In this study linear relation between intake and utilization of C18:2*n*-6 and C18:3*n*-3 in the body was affirmed. Similar relationship was shown by Nguyen *et al.* [2003], but for the subcutaneous and intramuscular fat. Moreover, the present results prove that efficiency of utilization of linoleic acid by growing pigs was similar to that presented by Kloarega *et al*. [2005] and much higher than reported by Flanzy *et al*. [1970], who also used the comparative slaughter technique. Nevertheless, shown in the present study as well as cited efficiency of utilization are both smaller than those given by Lizardo *et al*. [2002]. Moreover, all results cited above and relating to utilization of *n*-6 and *n*-3 fatty acids are not in accordance with those given by Chwalibog *et al*. [1992], who concluded that all digestible dietary lipids are stored in the body thus being fully (100%) utilized. However, these results were achieved using calorimetric data.

The results of this study as well as limited information from literature and discrepancy between the data relating efficiency of utilization of exogenous FA impede an explanation of mechanisms regulating relationships between supply and deposition in the pig's body of the fatty acids in question.

It could be concluded that increasing the intake of both linoleic and linolenic acids enhances their deposition in the pig's body. Efficiency of their utilization depends not only from amount consumed, but also from sources of their origin.

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