



Effect of linseed oil sediment in the diet of pigs on the growth performance and fatty acid profile of meat

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ABSTRACT - The objective of this investigation was to examine the influence of dietary linseed oil sediment on the growth performance and fatty acid composition in the muscle tissue of pigs. Sixty-eight crossbred Swedish Yorkshire × Norwegian Landrace pigs were allocated to two trials with two different levels of linseed oil sediment. Twenty-four pigs in Trial 1 were allotted into control 1 and experimental 1, of 12 animals each, and forty-four pigs in Trial 2 were allotted into control 2 and experimental 2, of 22 animals each. In both treatments, control and experimental groups were formed by animals analogous by origin, gender, weight, and condition score. Control pigs were fed identical diets *ad libitum* in both trials. The treated pigs were fed the same diet as control pigs, but vegetable oil was replaced by linseed oil sediment at a rate of 25 g kg⁻¹ (experimental group 1) in Trial 1 and 50 g kg⁻¹ (experimental group 2) in Trial 2. The results indicated that in both trials, vegetable oil replacement for linseed oil sediment had no significant influence on the growth rate of pigs, though a tendency was observed for a more rapid daily gain. Addition of linseed oil sediment to the diets increased the content of n-3 α -linolenic (C18:3n-3), eicosatrienoic (C20:3n-3), and eicosapentaenoic (C20:5n-3) acids and total n-3 polyunsaturated fatty acids (PUFA) and decreased the C18:2n-6/C18:3n-3 and n-6:n-3 ratios and the thrombogenic index of meat. Moreover, the addition of 50 g kg⁻¹ linseed oil sediment resulted in higher content of docosapentaenoic (C22:5n-3) fatty acid, total PUFA, and PUFA:SFA ratio. Supplementation of pig diets with linseed oil sediment increases the content of α -linolenic, eicosatrienoic, eicosapentaenoic, and docosapentaenoic fatty acids and total content of n-3 polyunsaturated fatty acids and have a positive effect by improving the polyunsaturated fatty acids:saturated fatty acids and n-6:n-3 fatty acid ratios in meat.

Key Words: oilseed, pig nutrition, saturated fatty acid, unsaturated fatty acid

Introduction

The cereal-based diet that is mostly used for pig feeding supplies a small amount of n-3 polyunsaturated fatty acids (PUFA). The aspiration to increase the n-3 PUFA content in pork requires a supply of n-3 PUFA from the diet (Nieto and Ros, 2012). Therefore, in the past few years, several studies have focused on the improvement of the nutritional value of pork. Pig feed has been oriented to a higher content of natural sources of PUFA to increase the tissue deposition of PUFA and to improve the health status of consumers (Boselli et al., 2008; Peiretti et al., 2015). Linseed is one of n-3 PUFA sources. Linseed contains from 18.9 to 27.0% protein and from 34.1 to 40.7% fat (Čolovic et al., 2016). Adding 5 or 10% of linseed in the diet of pigs increases

concentrations of α -linolenic acid and decreases n-6:n-3 ratio (Matthews et al., 2000).

Linseed oil in the diet throughout the fattening period increased the growth rate of pigs during the finishing period (Więcek et al., 2010). It also increased the content of C18:3 in neutral lipids and phospholipids in the *longissimus* muscle and *biceps brachii* muscle (Lu et al., 2008). The diet enriched with 3% linseed oil produced the highest level of α -linolenic acid (Nguyen et al., 2003). However, as an ingredient in pig rations, linseed will only be used as a secondary linseed oil product – as a meal or a cake – if its use is economically justified.

Corino et al. (2008) showed that inclusion of extruded linseed in the diet of pigs increased n-3 PUFA content in both *longissimus dorsi* muscle and backfat and decreased n-6:n-3 PUFA ratio. Inclusion of 13.4% ground linseed in the diet of pigs increased the content of linoleic and α -linolenic acids in the muscle tissue and backfat (Bečková and Václavková, 2010).

Storage of cold pressed oil in various containers produces sedimentation with a comparatively high content of fat, protein, and phospholipids. Obviously, linseed oil

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sediment has a lower content of α -linolenic fatty acid (C18:3n-3) compared with linseed or linseed oil, but at the same time also a lower content of linoleic fatty acid (C18:2n-6), harmful to health. However, there is no sufficient data on the use of this sediment in animal feeding. Therefore, the objective of this study was to investigate the effect of cold pressed linseed oil sediment in the diet of pigs on the growth performance and fatty acid composition in meat.

Material and Methods

This trial was carried out in accordance with the Directive 2010/63/EU of the European Parliament and the Council of 22 September, 2010 on the protection of animals used for scientific purposes.

Sixty-eight crossbred Swedish Yorkshire \times Norwegian Landrace pigs were used in two dietary experiments. Twenty-four pigs of 16.50 ± 0.47 kg body weight in Trial 1 were allotted into control 1 and experimental 1 groups, of 12 animals each, and forty-four pigs of 14.84 ± 0.45 kg body weight in Trial 2 were likewise allotted into control 2 and experimental 2 groups, of 22 animals each. In both trials, control and experimental groups were formed by animals analogous by origin, gender, weight, and condition score. The pigs of control groups were fed an identical diet which was adapted according to the physiological needs of pigs at different ages during growing and finishing periods (Table 1). In the finishing period, the dietary composition

was slightly changed by reducing the soybean meal content and increasing the amount of peas. The pigs of experimental groups were fed the same diet as control pigs, but vegetable oil was replaced by linseed oil sediment at a rate of 25 g kg^{-1} (experimental group 1) in Trial 1 and 50 g kg^{-1} (experimental group 2) in Trial 2 (Table 1).

The pigs in control and treated groups were fed *ad libitum* and raised in pens of 12.6 m^2 area with 11 or 12 pigs per pen. Feed intake was recorded by pen and average daily feed intake per pig per day and per kg gain was estimated by group of pigs. The finishing period ended when the pigs reached a final live weight of 110-120 kg.

The average air temperature and relative air humidity were recorded using LogTag Humidity&Temperature Recorder (NAXO-8). The temperature and humidity were, respectively, 17.7 ± 0.04 °C and $81.0 \pm 0.25\%$ (Trial 1) and 17.6 ± 0.04 °C and $81.5 \pm 0.24\%$ (Trial 2).

The chemical composition and nutritive value of the feeds were analysed according to the standard methods (AOAC, 1990). The analysis of nutritional value of vegetable and linseed oil sediment used in the pig feed showed that linseed oil sediment contained 4,51 times more α -linolenic (C18:3n-3) and 3,61 times less linoleic (C18:2n-6), fatty acids in comparison with vegetable oil (Table 2).

The growth rate at different periods was determined by individual weighing of pigs before the morning feeding at the start of the experiments, every month, and at the end of both experiments.

Table 1 - Composition and nutritive value of diets

Item	Trial 1				Trial 2			
			Group				Group	
			Control 1	Experimental 1			Control 2	Experimental 2
			Period				Period	
	Growing	Finishing	Growing	Finishing	Growing	Finishing	Growing	Finishing
Feedstuff								
Triticale (g kg^{-1})	400	410	400	410	400	410	400	410
Wheat (g kg^{-1})	360	350	349	339	360	350	324	314
Soybean meal (g kg^{-1})	100	50	96	46	100	50	96	46
Peas (g kg^{-1})	100	150	100	150	100	150	100	150
Vegetable oil (g kg^{-1})	10	10	-	-	10	10	-	-
Sediments of linseed oil (g kg^{-1})	-	-	25	25	-	-	50	50
Premix Lit358200-11/4915 (g kg^{-1})	30	30	30	30	30	30	30	30
Nutrient								
Dry matter (g kg^{-1})	869	867	871	869	869	867	874	872
Metabolizable energy (MJ)	13.3	13.3	13.5	13.5	13.3	13.3	13.8	13.8
Crude protein (g kg^{-1})	158.2	146.5	158.2	146.5	158.2	146.5	158.3	146.5
Lysine (g kg^{-1})	9.0	8.3	8.9	8.2	9.0	8.3	8.9	8.2
Methionine (g kg^{-1})	5.1	4.7	5.1	4.7	5.1	4.7	5.1	4.7
Threonine (g kg^{-1})	5.6	5.2	5.6	5.1	5.6	5.2	5.6	5.1
Tryptophane (g kg^{-1})	1.9	1.8	1.9	1.8	1.9	1.8	1.9	1.7
Fibre (g kg^{-1})	33.2	33.0	32.9	32.6	33.2	33.0	32.5	32.2
Calcium (g kg^{-1})	6.3	6.2	6.3	6.2	6.3	6.2	6.3	6.2
Phosphorus (g kg^{-1})	4.7	4.6	4.7	4.6	4.7	4.6	4.7	4.6

Table 2 - Composition of vegetable oil and linseed oil sediment

Item	Vegetable oil	Linseed oil sediment
Metabolizable energy (MJ kg ⁻¹)	35.0	28.0
Crude protein (g kg ⁻¹)	-	120.0
Fat (g kg ⁻¹)	1000.0	705.0
Fatty acids (g kg ⁻¹)		
Lauric (C12:0)	0.1	-
Myristic (C14:0)	3.9	0.2
Myristoleic (C14:1)	0.2	-
Pentadecanoic (C15:0)	0.8	-
Palmitic (C16:0)	102.3	28.5
Palmitoleic (C16:1n-7)	1.2	0.2
Hexadecenoic (C16:1n-9)	5.5	0.7
Margaric (C17:0)	1.4	0.5
Heptadecenoic (C17:1n-9)	1.0	0.3
Stearic (C18:0)	28.5	18.3
Oleic (C18:1n-9)	313.3	216.7
Elaidic (C18:1n-9 trans)	0.9	0.1
Linoleic (C18:2n-6)	425.2	117.9
Vaccenic (C18:1n-7)	29.3	-
α -linolenic (C18:3n-3)	68.9	310.6
γ -linolenic (C18:3n-6)	0.3	0.7
Arachidic (C20:0)	3.1	2.1
Eicosenoic (C20:1n-9)	3.8	4.8
Eicosadienoic (C20:2n-6)	0.4	0.3
Eicosatrienoic (C20:3n-3)	0.2	0.3
Behenic (C22:0)	3.8	1.3
Erucic (C22:1n-9)	0.5	0.3
Docosatetraenoic (C22:4n-6)	0.6	-
Lignoceric (C24:0)	2.0	0.7
Nervonic (C24:1n-9)	2.8	0.5

During control slaughtering in both trials, four analogous pigs were selected from each control and treated groups to sample collection of *longissimus lumborum* muscle for evaluation of the composition of intramuscular fat. Fatty acid composition in intramuscular fat, linseed oil sediment, and vegetable oil were determined by gas chromatography (GC-2010 SHIMADZU) after extraction by the method of Folch et al. (1957) and using transmethylation by the method of Christopherson and Glass (1969).

Atherogenic (AI), thrombogenic (TI) (Okrouhlá et al., 2013), and hypo/hypercholesterolemic (h/H) (Fernández et al., 2007) indices were calculated on the basis of fatty acid analysis data as follows:

$$AI = (C12:0 + 4 \times C14:0 + C16:0) / (MUFA + PUFA);$$

$$TI = (C14:0 + C16:0 + C18:0) / (0.5 \times MUFA + (0.5 \times n-6 \text{ PUFA} + 3 \times n-3 \text{ PUFA} + n-3/n-6 \text{ PUFA}));$$

$$h/H = [(sum \text{ of } C18:1n-9, C18:1n-7, C18:2n-6, C18:3n-6, C18:3n-3, C20:3n-6, C20:4n-6, C20:5n-3, C22:4n-6, C22:5n-3, \text{ and } C22:6n-3) / (sum \text{ of } C14:0 \text{ and } C16:0)];$$

in which, MUFA = monounsaturated fatty acids.

Processing of the data was performed using software Statistica (Data Analysis Software System, Version 7.0; StatSoft, Inc., Tulsa, OK, USA). The individual pig served as the experimental unit for all growth and polyunsaturated fatty acid indicators of meat. The statistical evaluation of

the results was performed using descriptive statistics and Student's t-test for independent samples. In the figures, all data are displayed as mean (\bar{x}) and standard error (SE) of the mean. The probability level of $P < 0.05$ was considered as statistically significant, whereas the differences of $0.05 < P < 0.10$ were considered as tendency to difference.

Results

The results of our study showed that growth performance was not influenced by treatment. However, in experimental group 1, a tendency was observed for a more rapid growth of pigs as the daily gain was 73 g higher ($P < 0.10$) than that of the pigs in control group 1 in Trial 1 (Table 3). The daily gain of the treated pigs in the growing (1.5-4 months of age) and finishing (over four months of age until slaughter) periods was higher by 60 and 87 g, respectively, than those of the control group, although no significant differences were found. In Trial 2, when a higher content (50 g kg⁻¹) of linseed oil sediment was used, the daily gain of the treated pigs in experimental group 2 was 73 g higher ($P < 0.10$) than that of control group 2 within the whole growing-finishing period. Meanwhile, in the growing period pigs of experimental group 2 gained 74 g ($P < 0.10$) daily more than pigs of control group 2. Treated pigs consumed more feeds than control pigs (Table 4). Within the whole experimental period, feed intake per kg gain of pigs of experimental groups 1 and 2 was lower, respectively, by 80 and 190 g in comparison with pigs of control groups 1 and 2. Although in Trial 1 no differences were found between the groups for the duration of the fattening time, there was a significant effect on this indicator (10.1 day less; $P < 0.025$) of treated pigs fed linseed oil sediment in Trial 2.

In both trials, 32 fatty acids were identified in intramuscular fat, of which nine were saturated (SFA), nine monounsaturated (MUFA), and 14 polyunsaturated (PUFA) (Table 5). In experimental group 1, there was a tendency for 0.02% lower capric (C10:0) ($P < 0.10$) and 1.6% lower palmitic (C16:0) ($P < 0.10$) SFA. Also, the decrease in the content of total saturated fatty acids (-1.63%; $P < 0.10$) was observed in experimental group 1. There was a tendency towards lower content of stearic (C18:0) fatty acid (-1.04%; $P > 0.10$) in the muscle of experimental group 2. In the muscle of pigs of experimental group 1, the contents of palmitoleic (C16:1n-7) and vaccenic (C18:1n-7) MUFA were, respectively by 0.61 ($P < 0.025$) and 0.57% ($P < 0.05$) lower, whereas the meat of pigs of experimental group 2 showed

a tendency towards a higher content of erucic (C22:1n-9) fatty acid (+0.01%; $P<0.10$).

Intramuscular fat of pigs of experimental group 1 contained more n-3 PUFA: the content of α -linolenic (C18:3n-3) was 1.36% or 3.4 times ($P<0.001$) higher, eicosatrienoic (C20:3n-3) was 0.18% or 2.3 times ($P<0.001$) higher, and eicosapentaenoic (C20:5n-3) was 0.1% or 1.7 times ($P<0.05$) higher in comparison with the pigs of control group 1. The intramuscular fat of pigs of experimental group 2 had higher contents of the following acids: α -linolenic (C18:3n-3), 1.14% or 2.4 times ($P<0.001$); eicosatrienoic (C20:3n-3), 0.18% or 2.3 times ($P<0.001$); eicosapentaenoic (C20:5n-3), 0.16% or 2.5 times ($P<0.001$); and docosapentaenoic (C22:5n-3), 0.18% or 1.8 times ($P<0.005$). In addition, there was a tendency observed for a higher content of docosahexaenoic (C22:6n-3) fatty acid (0.03%; $P<0.10$) in comparison with control group 2. Irrespective of the amount of linseed oil sediment in the diet of experimental groups 1 and 2, total n-3 PUFA in the intramuscular fat was significantly higher, from 2.24 to 2.34 times ($P<0.001$), in comparison with the pigs in control groups 1 and 2.

The results of our study showed a tendency for a higher content of linoleic (C18:2n-6) fatty acid (1.22%; $P<0.10$) in the intramuscular fat of pigs of experimental group 1, whereas in experimental group 2, there were tendencies observed for a higher content of docosadienoic (C22:2n-6) fatty acid (0.03%; $P<0.10$) and a lower content of docosatetraenoic (C22:4n-6) fatty acid (0.03%; $P<0.10$) in comparison with the pigs of control groups 1 and 2. Furthermore, it was found that the muscles of pigs of experimental group 1 had higher total PUFA (2.63%; $P<0.10$) and total PUFA in the muscle tissue of pigs of experimental group 2 was 2.26% higher ($P<0.05$). The PUFA:SFA ratio was higher and n-6:n-3 fatty acid ratio was lower in the intramuscular fat of experimental groups 1 and 2 by, respectively, 1.30 ($P<0.10$) - 1.31 times ($P<0.05$) and 2.11-2.14 times ($P<0.001$).

It was found that the thrombogenic index of meat of experimental groups 1 and 2 was, respectively, 17.1 ($P<0.001$) and 17.9% ($P<0.005$) lower in comparison with control groups 1 and 2. No significant decrease of the meat atherogenic index in both treated groups was observed in comparison with the control groups.

Table 3 - Growth rate of pigs

Item	Treatment 1			Treatment 2		
	Group		P-value ¹	Group		P-value ¹
	Control 1	Experimental 1		Control 2	Experimental 2	
	$\bar{x}\pm\text{SE}$	$\bar{x}\pm\text{SE}$		$\bar{x}\pm\text{SE}$	$\bar{x}\pm\text{SE}$	
Weight of pigs (kg)						
At the start of the experiment	16.56 \pm 0.688	16.44 \pm 0.680	0.910	14.85 \pm 0.601	14.83 \pm 0.681	0.977
At the end of growing period	62.12 \pm 2.680	66.47 \pm 2.843	0.282	58.72 \pm 2.251	64.26 \pm 2.636	0.121
At the end of finishing period	113.12 \pm 3.096	120.37 \pm 2.790	0.101	112.50 \pm 2.600	114.35 \pm 2.500	0.612
Average daily gain (g)						
In growing period	607 \pm 38.397	667 \pm 33.473	0.261	585 \pm 24.477	659 \pm 28.074	0.057
In finishing period	634 \pm 44.603	721 \pm 27.130	0.133	749 \pm 36.587	812 \pm 31.565	0.230
During the trial	621 \pm 28.690	694 \pm 19.500	0.052	665 \pm 25.023	728 \pm 20.359	0.061
Duration in fattening (days)	155.5 \pm 4.24	149.8 \pm 3.16	0.248	146.8 \pm 3.65	136.7 \pm 2.40	0.019

$\bar{x}\pm\text{SE}$ - mean \pm standard error.

¹ Significance level ($\alpha = 0.05$).

Table 4 - Feed intake

Item	Treatment 1		Treatment 2	
	Group		Group	
	Control 1	Experimental 1	Control 2	Experimental 2
Feed intake per pig per day (kg)				
In growing period	1.76	1.90	1.69	1.83
In finishing period	2.62	2.92	3.08	3.21
During the trial	2.21	2.41	2.37	2.45
Feed intake per kg/gain (kg)				
In growing period	2.90	2.85	2.89	2.78
In finishing period	4.13	4.05	4.11	3.95
During the trial	3.55	3.47	3.56	3.37

Table 5 - Fatty acid composition (%) of the *longissimus lumborum* muscle

Item	Treatment 1			Treatment 2		
	Group		P-value ¹	Group		P-value ¹
	Control 1	Experimental 1		Control 2	Experimental 2	
	$\bar{x} \pm \text{SE}$	$\bar{x} \pm \text{SE}$		$\bar{x} \pm \text{SE}$	$\bar{x} \pm \text{SE}$	
Capric (C10:0)	0.10±0.007	0.08±0.003	0.069	0.08±0.004	0.09±0.005	0.278
Lauric (C12:0)	0.11±0.031	0.08±0.010	0.354	0.08±0.007	0.07±0.007	0.791
Myristic (C14:0)	0.93±0.257	1.12±0.053	0.502	1.23±0.058	1.19±0.039	0.586
Pentadecanoic (C15:0)	0.04±0.025	0.01±0.010	0.300	0.04±0.005	0.01±0.013	0.111
Palmitic (C16:0)	24.64±0.475	23.04±0.567	0.073	25.38±0.447	24.53±0.339	0.181
Margaric (C17:0)	0.33±0.060	0.39±0.009	0.644	0.25±0.099	0.23±0.029	0.907
Stearic (C18:0)	11.79±0.179	11.60±0.235	0.548	12.36±0.370	11.32±0.413	0.108
Arachidic (C20:0)	0.19±0.006	0.20±0.027	0.685	0.22±0.012	0.19±0.017	0.230
Behenic (C22:0)	0.07±0.024	0.07±0.012	0.855	0.04±0.004	0.04±0.005	0.705
SFA	38.20±0.575	36.57±0.429	0.064	39.67±0.778	37.67±0.772	0.119
Myristoleic (C14:1n-7)	0.01±0.008	0.00±0.000	0.356	0.00±0.000	0.01±0.008	0.356
Palmitoleic (C16:1n-7)	3.04±0.102	2.43±0.152	0.016	2.75±0.142	2.77±0.087	0.920
Hexadecenoic (C16:1n-9)	0.25±0.006	0.30±0.030	0.189	0.21±0.019	0.22±0.014	0.838
Heptadecenoic (C17:1n-9)	0.17±0.012	0.19±0.020	0.426	0.21±0.015	0.25±0.025	0.218
Vaccenic (C18:1n-7)	4.06±0.172	3.49±0.111	0.033	3.56±0.137	3.61±0.134	0.774
Oleic (C18:1n-9)	41.53±1.043	41.93±1.346	0.820	41.18±0.829	40.55±0.355	0.509
Elaidic (C18:1n-9 trans)	0.18±0.019	0.18±0.015	1.00	0.20±0.034	0.21±0.012	0.893
Eicosenoic (C20:1n-9)	0.79±0.034	0.75±0.045	0.501	0.84±0.030	0.81±0.056	0.623
Erucic (C22:1n-9)	0.06±0.013	0.08±0.018	0.526	0.05±0.003	0.06±0.005	0.067
MUFA	50.07±0.907	49.33±1.176	0.636	48.99±0.979	48.47±0.433	0.644
Linoleic (C18:2n-6)	6.90±0.288	8.12±0.552	0.099	7.51±0.339	8.05±0.479	0.397
Linolelaidic (C18:2n-6 trans)	0.08±0.011	0.07±0.016	0.900	0.05±0.006	0.07±0.009	0.114
Octadecadienoic (C18:2n-6 cis,trans)	0.04±0.021	0.01±0.010	0.317	0.00±0.000	0.03±0.025	0.356
γ -linolenic (C18:3n-6)	0.05±0.018	0.08±0.016	0.324	0.07±0.007	0.08±0.016	0.585
α -linolenic (C18:3n-3)	0.57±0.029	1.93±0.085	<0.001	0.81±0.061	1.95±0.142	<0.001
Eicosadienoic (C20:2n-6)	0.28±0.025	0.29±0.028	0.747	0.29±0.004	0.28±0.017	0.675
Eicosatrienoic (C20:3n-3)	0.14±0.017	0.32±0.042	<0.001	0.14±0.015	0.32±0.017	<0.001
Eicosatrienoic (C20:3n-6)	0.19±0.025	0.19±0.041	1.00	0.16±0.017	0.18±0.016	0.420
Arachidonic (C20:4n-6)	1.26±0.260	0.97±0.240	0.451	0.83±0.109	0.81±0.078	0.886
Eicosapentaenoic (C20:5n-3)	0.14±0.016	0.24±0.031	0.028	0.11±0.007	0.27±0.029	0.001
Docosadienoic (C22:2n-6)	0.03±0.017	0.06±0.011	0.154	0.01±0.008	0.04±0.015	0.081
Docosatetraenoic (C22:4n-6)	0.18±0.039	0.10±0.025	0.166	0.13±0.014	0.10±0.011	0.093
Docosapentaenoic (C22:5n-3)	0.31±0.037	0.44±0.075	0.162	0.23±0.013	0.41±0.030	0.002
Docosahexaenoic (C22:6n-3)	0.14±0.025	0.09±0.019	0.196	0.07±0.014	0.10±0.007	0.072
PUFA	10.27±0.619	12.90±1.056	0.075	10.41±0.481	12.67±0.745	0.043
Unknown	1.47±0.257	1.20±0.297	0.521	0.94±0.074	1.19±0.098	0.090
n-6 PUFA	8.99±0.585	9.88±0.896	0.434	9.05±0.467	9.63±0.595	0.470
n-3 PUFA	1.29±0.049	3.02±0.196	<0.001	1.36±0.069	3.05±0.158	<0.001
n-6/n-3	6.99±0.32	3.27±0.18	<0.001	6.68±0.44	3.16±0.07	<0.001
Linoleic C18:2n-6/ α -linolenic C18:3n-3	12.39±1.19	4.22±0.28	<0.001	9.38±0.75	4.15±0.19	<0.001
PUFA/SFA	0.27±0.02	0.35±0.03	0.056	0.26±0.01	0.34±0.03	0.040
MUFA/SFA	1.31±0.04	1.35±0.03	0.478	1.24±0.05	1.29±0.03	0.394
MUFA/PUFA	4.94±0.69	3.92±0.82	0.107	4.75±0.62	3.86±0.46	0.06
Atherogenic index	0.47±0.024	0.44±0.013	0.353	0.51±0.019	0.48±0.013	0.235
Thrombogenic index	1.11±0.029	0.92±0.020	0.001	1.17±0.032	0.96±0.035	0.004
Hypo/hypercholesterolemic index	2.01±0.07	2.25±0.08	0.060	1.92±0.06	2.05±0.05	0.161

SFA - saturated fatty acids; PUFA - polyunsaturated fatty acids; MUFA - monounsaturated fatty acids; $\bar{x} \pm \text{SE}$ - mean \pm standard error.¹Significance level ($\alpha = 0.05$).

Discussion

The results of our study showed that in both trials, no significant difference in the growth rate between the pigs of the treated and control groups was found and this is in agreement with the findings of Morel et al. (2006), Nurnberg et al. (2011), and Okrouhlá et al. (2013), who used linseed or linseed oil in the diet. However, in our trials, a tendency

was observed for a more rapid daily gain and lower feed intake per kg gain and significantly shorter duration of fattening time in the pigs treated by linseed oil sediment. Higher feed conversion value in barrows fed linseed was also determined in the findings of Okrouhlá et al. (2013).

The analysis of amino acids showed that palmitic (C16:0) and stearic (C18:0) fatty acids were dominant in the meat of both control and treated pigs and there was

no difference for these fatty acids between the groups. A similar result for the stearic (C18:0) fatty acid was reported by Okrouhlá et al. (2013), although the same author, in contrast to our findings, reported a significantly lower content of palmitic (C16:0) fatty acid in the meat of pigs fed linseed.

It was found that the treatment of experimental group 1 resulted in significantly lower content of palmitoleic (C16:1n-7) and vaccenic (C18:1n-7) MUFA. Okrouhlá et al. (2013) also reported lower content of palmitoleic (C16:1n-7) fatty acid in the study with linseed, but as MUFA can be synthesized in the pig body, their contents in meat are not so important as those of PUFA (Enser et al., 2000; Kralik et al., 2010). Bečková and Václavková (2010) and Okrouhlá et al. (2013) indicated that linseed in the diet of pigs decrease total MUFA; however, no similar results were found in our study. The current results revealed that the MUFA:PUFA ratio decreased by 1.02-0.89 units in the meat of experimental groups 1 and 2, thereby improving the quality of pork. This agrees with the results reported by Okrouhlá et al. (2013), who also showed that dietary linseed supplement decreased the MUFA:PUFA ratio.

Pigs fed standard compound feed produce “unhealthy” meat due to improper n-6:n-3 ratio in it. This is because of the existing regularity that the composition of polyunsaturated fatty acids in pig feeds influences the PUFA composition in meat (Nguyen et al., 2003). A conventional feed for fattening pigs contains insufficient amount of n-3 fatty acids and has improper n-6:n-3 and PUFA:SFA ratios (Kralik et al., 2010). As linoleic (C18:2n-6) and α -linolenic (C18:3n-3) PUFA cannot be synthesized by porcine organism, the contents of these fatty acids in meat are dependent on their contents in the feed (Okrouhlá et al., 2013). As expected, the results of our trials indicated that there was a significant increase in α -linolenic (C18:3n-3), eicosatrienoic (C20:3n-3), and eicosapentaenoic (C20:5n-3) fatty acids in the meat of both treated groups. This agrees with the findings of Riley et al. (2000), Enser et al. (2000), Rey et al. (2004), and Kralik et al. (2010), who also indicated that dietary linseed or linseed oil supplement resulted in a significant increase of the above-mentioned fatty acids.

Our study resulted in significantly higher content of docosapentaenoic (C22:5n-3) fatty acid and this confirms the findings of Enser et al. (2000), who supplemented the pig diet with linseed oil. However, it should be noted that no significant differences for the docosahexaenoic (C22:6n-3) fatty acid were found in our study between the treated and control groups, notwithstanding the intake of linseed oil sediment. This agrees with the findings of Riley et al. (2000), Wood et al. (2003), and Václavková and

Bečková (2007), who supplemented pig feed with linseed or linseed oil, but contradicts the findings of Cunnane et al. (1990), Enser et al. (2000), and Kralik et al. (2010), who reported a significant increase of C22:6n-3 in the meat.

The meat of the treated pigs contained a significantly higher amount of α -linolenic (C18:3n-3), eicosatrienoic (C20:3n-3), and eicosapentaenoic (C20:5n-3) fatty acids. Also, the meat from experimental group 2 had a higher content of docosapentaenoic (C22:5n-3) fatty acid. The total of eicosapentaenoic (C20:5n-3) and docosahexaenoic (C22:6n-3) fatty acids, very important for human nutrition, in the meat of experimental group 2 was 0.37 versus 0.18% in the control group 2. This agrees with the finding of Enser et al. (2000), Nuernberg et al. (2005), and Okrouhlá et al. (2013) that, in the body of non-ruminants, long chain polyunsaturated C20-C22 n-3 fatty acids are synthesized from the α -linolenic (C18:3n-3) fatty acid found in the feed. In other words, our study showed that addition of linseed oil sediment in the pig diets had a positive effect on these fatty acids.

The results of our study indicated that the higher the dietary content of α -linolenic (C18:3n-3) fatty acid, the higher the content of the same fatty acid and other n-3 fatty acids in pork, which agrees with the findings of Rentfrow et al. (2003) and Okrouhlá et al. (2013). Nuernberg et al. (2005) and Václavková and Bečková (2007) reported that supplementation of pig diets with linseed or linseed oil resulted in lower total content of n-6 fatty acids and specifically of arachidonic (C20:4n-6) fatty acid; however, our findings in both experimental groups do not confirm this.

Enser et al. (1996) and Wood et al. (2003) observed that PUFA:SFA ratio in the meat of pigs fed conventional compound feeds is lower than that recommended for human nutrition (over 0.4). In our study with linseed oil sediment, this ratio in treated groups increased by 0.08 unit ($P < 0.05$ and $P < 0.10$). Similar results were reported by Nurnberg et al. (2011) and Okrouhlá et al. (2013), who supplemented pig feeds with linseed or linseed oil.

Additionally, Wood and Enser (1997) and Enser et al. (2000) indicated that the unusual n-6:n-3 fatty acid ratio in pork is above 10. The recommended ratio for human nutrition should be 4-5 or even lower (Okrouhlá et al. 2013). In our study, this ratio in the meat of the treated pigs, if compared with that of the control pigs, was 2.11-2.14 times lower and was equal to 3.16-3.27. This is in agreement with the findings of Sheard et al. (2000) and Wood et al. (2003), who indicated that dietary linseed supplement reduced the ratio to 4 or even less. Rey et al. (2004) and Kralik et al. (2010) also reported that supplementation of pig diets with

linseed oil had a significant effect on the ratio in pork, making it more suitable for human nutrition.

Pork contains rather high content of linoleic (C18:2n-6) fatty acid, which has a negative influence on the C18:2: C18:3 ratio important for human nutrition. In our study, this ratio decreased from 2.3 in Trial 2 to 2.9 in Trial 1 in the treated groups as compared with the control groups and was very close to the recommended value (4.0) in a healthy food (Wood et al., 2003).

It was found that in experimental groups 1 and 2, the thrombogenic index was significantly lower. The same conclusion was reached by Okrouhlá et al. (2013), who also reported lower thrombogenic index in the trials with linseed. Furthermore, as it was shown in our study, the atherogenic index of meat was by 0.03 times lower in experimental groups 1 and 2, but the differences were insignificant. Analogous data were reported by Okrouhlá et al. (2013) in the study of linseed in pig diets.

Conclusions

Supplementation of pig diets with linseed oil sediment has no significant influence on the growth performance of pigs; however, it increases the content of α -linolenic, eicosatrienoic, eicosapentaenoic, docosapentaenoic fatty acids and total content of n-3 polyunsaturated fatty acids and have a positive effect by improving the polyunsaturated fatty acids/saturated fatty acids and n-6:n-3 fatty acid ratios in meat and reducing the thrombogenic index of pork.

References

- AOAC - Association of Official Analytical Chemistry. 1990. Official methods of analysis. 15th ed. AOAC International, Arlington, VA.
- Bečková, R. and Václavková, E. 2010. The effect of linseed diet on carcass value traits and fatty acid composition in muscle and fat tissue of fattening pigs. *Czech Journal of Animal Science* 55:313-320.
- Boselli, E.; Pacetti, D.; Lucci, P.; Di Lecce, G. and Frega, N. G. 2008. Supplementation with high-oleic sunflower oil and α -tocopheryl acetate: Effects on pork meat lipids. *European Journal of Lipid Science and Technology* 110:381-391.
- Christopherson, S. W. and Glass, R. L. 1969. Preparation of milk fat methyl esters by alcoholysis in an essentially nonalcoholic solution. *Journal of Dairy Science* 52:1289-1290.
- Corino, C.; Musella, M. and Mouro, J. 2008. Influence of extruded linseed on growth, carcass composition, and meat quality of slaughtered pigs at one hundred ten and one hundred sixty kilograms of liveweight. *Journal of Animal Science* 86:1850-1860.
- Cunnane, S. C.; Stitt, P. A.; Ganguli, S. and Armstrong, J. K. 1990. Raised omega-3 fatty acid levels in pigs fed flax. *Canadian Journal of Animal Science* 70:251-254.
- Čolovic, D.; Berenji, J.; Levart, A.; Levic, J.; Salobir, J.; Pezo, L. and Čolovic, R. 2016. Nutritional characteristics of seeds of eighteen linseed (*Linum humile* Mill.) cultivars from Serbia. *Zemdirbyste-Agriculture* 103:175-182.
- Enser, M.; Hallett, K.; Hewitt, B.; Fursey, G. A. J. and Wood, J. D. 1996. Fatty acid content and composition of English beef, lamb and pork at retail. *Meat Science* 42:443-456.
- Enser, M.; Richardson, R. I.; Wood, J. D.; Gill, B. P. and Sheard, P. R. 2000. Feeding linseed to increase the n-3 PUFA of pork: fatty acid composition of muscle, adipose tissue, liver and sausages. *Meat Science* 55:201-212.
- Fernández, M.; Ordóñez, J. A.; Cambero, I.; Santos, C.; Pin, C. and de la Hoz, L. 2007. Fatty acid compositions of selected varieties of Spanish ham related to their nutritional implications. *Food Chemistry* 101:107-112.
- Folch, J.; Less, M. and Sloane-Stanley, G. H. 1957. A simple method for isolation and purification of total lipids from animal tissues. *Journal of Biological Chemistry* 226:497-509.
- Kralik, G.; Margeta, V.; Suchy, P. and Strakova, E. 2010. Effects of dietary supplementation with rapeseed and linseed oil on the composition of fatty acids in porcine muscle tissue. *Acta Veterinaria Brno* 79:363-367.
- Lu, P.; Zhang, L. Y.; Dyin, J. D.; Everts, A. K. R. and Li, D. F. 2008. Effects of soybean oil and linseed on fatty acid composition of muscle lipids and coked pork flavour. *Meat Science* 80:910-918.
- Matthews, K. R.; Homer, D. B.; Thies, F. and Calder, P. C. 2000. Effect of whole linseed (*Linum usitatissimum*) in the diet of finishing pigs on growth performance and on the quality and fatty acid composition of various tissues. *British Journal of Nutrition* 83:637-643.
- Morel, P. C. H.; McIntosh, J. C. and Janz, J. A. M. 2006. Alteration of the fatty acid profile of pork by dietary manipulation. *Asian-Australasian Journal of Animal Sciences* 19:431-437.
- Nguyen, L. Q.; Nuijens, M. C. G. A.; Everts, H.; Salden, N. and Beynen, A. C. 2003. Mathematical relationships between the intake of n-6 and n-3 polyunsaturated fatty acids and their contents in adipose tissue of growing pigs. *Meat Science* 65:1399-1406.
- Nieto, G. and Ros, G. 2012. Modification of fatty acid composition in meat through diet: effect on lipid peroxidation and relationship to nutritional quality – a review. p.239-258. In: *Lipid peroxidation*. Angel Catala, ed. Available at: <<http://cdn.intechopen.com/pdfs-wm/38460.pdf>>. Accessed on: Dec. 10, 2016.
- Nuernberg, K.; Fischer, K.; Nuernberg, G.; Kuchenmeister, U.; Klosowska, D.; Eliminowska-Wenda, G.; Fiedler, I. and Ender, K. 2005. Effects of dietary olive and linseed oil on lipid composition, meat quality, sensory characteristics and muscle structure in pigs. *Meat Science* 70:63-74.
- Nurnberg, K.; Nurnberg, G.; Dannenberger, D.; Hagemann, L. and Paulke, T. 2011. Effect of extruded linseed on growth and lipids of muscle and back fat in pigs. *Fleischwirtschaft* 91:88-92.
- Okrouhlá, M.; Stupka, R.; Čítek, J.; Šprysl, M. and Brzobohatý, L. 2013. Effect of dietary linseed supplementation on the performance, meat quality, and fatty acid profile of pigs. *Czech Journal of Animal Science* 58:279-288.
- Peiretti, P. G.; Gai, F.; Brugiapaglia, A.; Mussa, P. P. and Meineri, G. 2015. Fresh meat quality of pigs fed diets with different fatty acid profiles and supplemented with red wine solids. *Food Science and Technology* 35:633-642.
- Rey, A.; Lopez-Bote, C. J.; Kerry, J. P.; Lynch, P. B.; Buckley, D. J. and Morrissey, P. A. 2004. Modification of lipid composition and oxidation in porcine muscle and muscle microsomes as affected by dietary supplementation of n-3 with either n-9 or n-6 fatty acids and alpha-tocopheryl acetate. *Animal Feed Science and Technology* 113:223-238.

- Rentfrow, G.; Sauber, T. E.; Allee, G. L. and Berg, E. P. 2003. The influence of diets containing either conventional corn with choice with grease, high oil corn, or high oil high oleic corn on belly/bacon quality. *Meat Science* 64:459-466.
- Riley, P. A.; Enser, M.; Nute, G. R. and Wood, J. D. 2000. Effects of dietary linseed on nutritional value and other quality aspects of pig muscle and adipose tissue. *Animal Science* 71:483-500.
- Sheard, P. R.; Enser, M.; Wood, J. D.; Nute, G. R.; Gill, B. P. and Richardson, R. I. 2000. Shelf life and quality of pork and pork products with a raised $n - 3$ PUFA. *Meat Science* 55:213-221.
- Václavková, E. and Bečková, R. 2007. Effect of linseed in pig diet on meat quality and fatty acid content. *Archiv Tierzucht* 50:144-151.
- Więcek, J.; Rekiel, A. and Skomiał, J. 2010. Effect of feeding level and linseed oil on some metabolic and hormonal parameters and on fatty acid profile of meat and fat in growing pigs. *Archiv Tierzucht* 53:37-49.
- Wood, J. D.; Richardson, R. I.; Nute, G. R.; Fisher, A. V.; Campo, M. M.; Kasapidou, E.; Sheard, P. R. and Enser, M. 2003. Effects of fatty acids on meat quality: a review. *Meat Science* 66:21-32.