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Medium-chain fatty acids as feed supplements for weaned piglets

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Summary

The aim of this experiment was to examine the effect of medium- and short-chain fatty acids (Selacid preparation) on piglets' health, body weight gains, changes in intestinal microflora and intestinal wall morphology. The amount of aerobic and anaerobic bacteria, yeasts and moulds, the acidity and the content of volatile fatty acids (VFA) of digesta were estimated. The effect of these acids was compared with the effect of the traditional acidifier i.e. fumaric acid. The experiment involved 183 piglets allocated to 3 experimental groups, 6 litters in each. Group I (control) was fed with the standard mixture with no supplement. Group II received the same mixture supplemented with 1.5% of fumaric acid and group III received feed mixture supplemented with 0.5% of the Selacid preparation. Experiment was completed at the 84th day of the piglets' life.

Selacid improved the piglets' body weight gains when compared to the control group (283 and 268 g, respectively) but this difference was not significant. Body weight gains of piglets receiving fumaric acid (269 g) did not differ from the control ones (268 g). Selacid had a moderate antibacterial effect: it lowered the number of Clostridium in small intestine digesta by 33%. Fumaric acid significantly lowered the amount of Escherichia coli. Acids had no effect on yeast and mould population.

Piglets receiving fumaric acid had higher intestinal villi than those receiving Selacid and the control ones (302, 257 and 233 µm, respectively) but that had no effect on their body weight gains. The content of volatile fatty acids and the acidity of intestinal digesta were also similar in all groups.

It is concluded that medium-chain fatty acid contained in the Selacid preparation can improve piglets' health and performance though this improvement is not related to changes in microflora or the structure of intestine mucosa in the scope of this research.

Keywords: medium-chain fatty acids, piglets' performance, intestinal morphology, intestinal microflora

Medium- and short-chain fatty acids, thanks to their antimicrobial activity (5, 27), could be good alternatives to feed antibiotics banned in the European Union countries. According to Dierick et al. (5) medium-chain fatty acids (MCFA) include: caproic (C_6), caprylic (C_8), capric (C_{10}) and lauric (C_{12}) acids. Mechanism of their antimicrobial action is not fully understood but probably they diffuse into bacterial cells in an undissociated form and dissociate within the protoplasm, leading to intracellular acidification. A lower intracellular pH can lead to inactivation of intracellular enzymes (30) and inhibition of amino acid transport (8, 17). The bacterial resistance against medium-chain fatty acids and monoacylglycerols was also found to be relatively less developed compared with other common antimicrobials. Probably this was due to multiple mechanisms by which fatty acids kill bacteria (13).

MCFA could also be a good energy source especially for young animals when compared with longer-chain fatty acids. They are readily absorbed through the stomach and the intestine, and they are transported directly to the liver *via* the portal vein and thus do not pass the adipose tissue before hepatic disposal (15). There they are oxidized in the mitochondria to CO_2, providing a rapidly available energy source for the mammalian newborn (14).

Other organic acids are also used as feed supplements, preventing diarrhoea among piglets (29). They can modify microbial population in the digestive tract (6) and improve the performance of piglets. One of them is fumaric acid which can improve body weight gains and the gain to feed ratio of weaned piglets (24). According to Skřivanová and Morounek (27) its beneficial effect is probably related to factors other than their direct antimicrobial action.

Organic acids can also affect the morphology of the intestinal wall. Gálfi and Bokori (10) observed that butyrate increased the length of ileal microvilli and depth of cecal crypts. Also Piva et. al. (18), feeding piglets with precursors of butyric acid, found an improvement in animal growth and a reduction of cecal crypt depth. On the other hand, Biagi et al. (2) did not find any significant effect of gluconic acid on intestinal villi or crypts but they collected samples 6 weeks after weaning, i.e. from piglets with an almost fully developed digestive system. In available literature we have not found information on the effect of MCFA on the morphology of the intestinal wall.

The aim of this experiment was to investigate the effect of a mixture of medium-chain fatty acids or fumaric acid on piglets' growth performance, intestinal microflora and the morphology of the intestinal wall.

Material and methods

The experiment was performed on 183 piglets originating from Polish Landrace × Polish Large White sows mated with Pietrain × Hampshire boar. Piglets were allocated to three experimental groups, each consisting of 6 litters. All piglets received the standard wheat – soybean feed mixture (tab. 1) available *ad libitum* from the 7th day of life. Group I (control) received feed with no supplement, group II was fed feed with 1.5% fumaric acid. Group III received feed supplemented with 0.5% of Selacid Green Growth (Selko B.V., Tilburg, The Netherland), which is a mixture of short-chain and medium-chain (i.e. caproic (C_6), caprylic (C_8), capric (C_{10}) and lauric (C_{12})) acids. Piglets had free access to water. The experiment lasted until the 84th day of life, and the body weight of individual piglets was checked at the 1st, 35th, 56th and 84th days of life. Their conditions were observed, feed intake of each litter was measured and feed utilization was calculated.

Between the 52nd and 58th day of life 18 piglets (6 from each treatment) were slaughtered. Fragments of small intestines were extracted to determine the height and width of villi and to measure the crypts. The extracted material was fastened to polystyrene plates and fixed in 10% buffered solution of formalin. Longitudinal strips, two for each specimen were taken. They were transformed using the Shandon Company tissue processes and embedded in paraffin. Sections of 3 mm were made from the paraffin blocks and dyed using the hematoxylin-eosin method. Only the villi with uniform fibrovascular stroma were used in the measurement i.e. those cut along the longitudinal axis. For each villi 3 longitudinal and 3 transversal measurements were made using the Zeiss Anxioscop microscope and CDD ZVS-47DE camera.

Microbiological tests were made on caecum and small intestine digesta. The number of aerobic bacteria, especially *Escherichia coli* and anaerobic ones, especially *Clostridium sp.*, was determined. The presence of yeasts and moulds was also estimated. The tests were made with the plate methods using agar medium by bioMerieux, according to Polish Standards (19-23).

Tab. 1. The composition and nutritive value of the feed mixture (%)

Components	Control	Fumaric acid	Selacid
Wheat, ground			
Barley, ground	10.00	10.00	10.00
Triticale	10.00	8.00	9.60
Soybean meal	25.00	25.00	25.00
Milk powder	3.00	3.00	3.00
Dried whey	5.00	5.00	5.00
Rapeseed oil	1.00	1.00	1.00
Dicalcium phosphate	0.80	0.80	0.80
Limestone	1.2	1.10	1.10
Salt	0.25	0.25	0.25
L-lysine	0.10	0.10	0.10
DL-Methionine	0.15	0.15	0.15
Premix*	0.5	0.5	0.5
Fumaric acid	–	1.5	–
Selacid	–	–	0.5
Content of nutrients in 1 kg of mixture			
Metabolizable energy (MJ)	12.88	12.74	12.89
Crude protein (g)	200.0	197.0	198.0
Crude fat (g)	26.0	23.3	22.4
Crude fibre (g)	29.1	26.2	27.6
N-free extractives (g)	589.5	590.8	592.1
Lysine (g)	10.5	10.5	10.5
Methionine + cystine (g)	7.65	7.59	7.63
Calcium (g)	9.44	9.73	9.73
Phosphorus (g)	6.69	5.88	5.92

Explanations: * – premix composition: vitamin: A – 2 700 000 IU; D3 – 400 000 IU; E – 8.0 g; K3 – 0.5 g ; B1 – 0.5 g; B2 – 0.8 g; B6 – 0.8 g; B12 – 0.008 g; pantothenic acid – 2.8 g; choline cloride – 70 g; folic acid – 0.2 g; nicotinic acid – 5.0 g; magnesium – 10 g; manganese – 12 g; iodine – 0.1 g; zinc – 30 g; iron – 20 g; copper – 32 g; cobalt – 0.06 g; selenium – 0,04 g; complete limestone to 1000 g

The content of nutrients in feed mixture was determined according to AOAC (1) methods and that of volatile fatty acids was estimated using the VARIAN 3400 gas chromatograph. The pH of digesta was measured with the CP – 411 pH-meter equipped with the Metron 12-01 electrode.

The data obtained were analyzed statistically by ANOVA and the significance of differences was examined using Duncan's test (Statistica Software, 5.1).

Results and discussion

The supplement of fumaric acid and Selacid reduced amount of dead and culled piglets from about 17% in control to about 14% in both experimental groups (tab. 2). Probably this was due to antimicrobial effect of organic acids. These findings are in accordance with the results of Skřivanová and Marounek (26), who

Tab. 2. Indices of piglet performance

Indices	Control	Fumaric acid	Selacid	SEM
No of litters in treatment	6	6	6	–
No of born piglets in treatment	58	63	62	–
Average No of piglets born per litter	9.66	10.50	10.33	–
Average No of piglets weaned per litter	9.00	10.33	9.33	–
Average No of piglets at day 84 per litter	8.00	9.00	8.83	–
Dead and culled piglets (%)	17.51	14.28	14.51	–
Body weight (kg) on days of life:				
1st	1.66	1.69	1.70	0.02
35th	8.14	8.19	8.55	0.13
56th	10.91	10.83	11.67	0.19
84th	23.88	24.05	25.21	0.41
Average daily gain (g) in periods of life:				
1st-35th day	191	191	201	3.71
35th-56th day	132ab	126a	148b	4.21
56th-84th day	463	472	483	10.85
35th-84th day	321	324	340	6.97
1st-84th day	268	269	283	4.93
Feed conversion ratio (kg/kg) in periods of life:				
1st-35th day	0.103	0.101	0.095	0.002
35th-56th day	2.13	2.17	1.54	0.148
56th-84th day	2.21	2.06	2.04	0.056
35th-84th day	2.11	2.02	1.95	0.048
1st-84th day	1.48	1.44	1.37	0.028

Explanations: a, b – mean values in the same row with different letters differ significantly at $P \leq 0.05$

found that caprylic acid added to feed for rabbits lowered mortality from 16.7 to 0%. Also fumaric acid lowered bacterial population in the digestive tract of piglets in the experiment of Gabert et al. (9), and reduced post-weaning diarrhoea in the experiment of Tsiloyiannis et al. (29).

Selacid improved body weight gains of piglets between the 35th day of life and the end of the experiment but the differences between Selacid (340 g) and the control group (321 g) were not significant. Similar results were obtained for the whole experimental period (283 g and 268 g, respectively). Also Cera et al. (3) found increased body weight gains of weaned piglets when feed was supplemented with capric and caprylic acids. Dierick et al. (4) obtained appreciable body weight gains of piglets fed seeds (*Cuphea*) rich in medium chain fatty acids. In the current experiment the supplement of fumaric acid did improve the results (269 g) in comparison with control (268 g). This accords with the results of Giesting and Easter (11), who also found no improvement in nutrient digestibility or better gains in young pigs fed diets supplemented with this acid. On the other hand, Lawlor et al. (12) found improved post-weaning performance of piglets fed with fumaric acid. Feed supplemented with fumaric acid in the current experiment was not eaten as readily as the other two but differences were not significant in any part of the experiment.

The medium-chain fatty acids preparation had only moderate antimicrobial activity (tab. 3). It lowered the amount of *Clostridium* in small intestine digesta by about 33% when compared to the control but this difference was not significant. Also Skřivanová et al. (28) found two strains of *Clostridium* to be susceptible to capric acid. Fumaric acid significantly lowered *E. coli* population (2.25) in comparison with the control (4.70). Similar results were obtained by Tsiloyiannis et al (29).

Acids did not lower the amount of yeasts. Their population in the group receiving Selacid was even slightly higher than that in the control animals. On the other hand Selacid lowered the amount of moulds but neither of these differences was statistically significant.

Organic acid can increase the villus height and also decrease crypt depth thus increasing the absorptive capacity of the intestinal epithelium. Such an effect was found by Gálfi and Bokori (10) in piglets receiving sodium butyrate. Similar results were obtained in the experiment on chickens by Pelicano et al. (16). In the current experiment (tab. 4) piglets receiving fumaric acid

Tab. 3. Microorganisms in small intestine chyme (Log_{10} CFU/1 g)

Number of microorganisms	Control	Fumaric acid	Selacid	SEM
Aerobic bacteria	7.21	6.59	7.18	0.280
– *E. coli*	4.70a	2.25b	4.03a	0.407
Anaerobic bacteria	6.61	6.37	7.35	0.341
– *Clostridium*	3.68	2.44	2.55	0.277
Candida albicans	2.70	2.78	3.40	0.258
Moulds	3.42	3.22	2.94	0.131
Moulds and micos	3.68	3.39	3.61	0.121

Explanations: a, b, – mean values in the same row with different letters differ significantly at $P \leq 0.05$

Tab. 4. Morphological characteristics of the small intestine epithelium

Estimated data	Control	Fumaric acid	Selacid	SEM
Villus height (µm)	233	302	257	15.51
Villus width (µm)	116	122	128	3.30
Crypt depth (µm)	280	307	294	10.58
Villus height/crypt depth	0.835	0.990	0.920	0.04

Tab. 5. The content of volatile fatty acids (μmol/1 g) in piglets' ileum chyme

VFA	Control	Fumaric acid	Selacid	SEM
Acetic	3.860	3.420	2.330	0.82
Propionic	0.666	0.287	0.444	0.09
Butyric	0.371	0.298	0.088	0.07
Isobutyric	0.160	0.153	0.090	0.02
Valeric	0.041	0.060	0.008	0.01
Isovaleric	0.078	0.050	0.033	0.01
Total acids	5.174	4.272	3.000	0.92

Tab. 6. The acidity of digesta in the stomach and in different parts of the small intestine

pH	Control	Fumaric acid	Selacid	SEM
Stomach	2.74	2.34	2.22	0.174
Small intestine				
– beginning	5.26	5.72	5.49	0.222
– middle	5.89	5.53	5.69	0.101
– end	5.66	5.42	5.65	0.132
– caecum	5.36	5.12	5.43	0.082

had higher villi then those receiving Selacid and the control ones (302; 257 and 233 μm, respectively). Higher villi in group II had no positive effect on body weight gains of the piglets.

There were no significant differences between the groups in the levels of volatile fatty acids in the small intestine content though the level of acetic acid in the digesta of pigs receiving Selacid was slightly lower than that of the other two groups (tab. 5). Also the pH of digesta was similar for all piglet groups except for a lower pH in the duodenum of animals from the control group (tab. 6). Similarly small differences in the pH of the content of different parts of the digestive tract were found by Franklin et al. (7) though pH values found in the current experiment were slightly lower. The lack of significant differences in the measurements of intestinal contents accords with the statement of Risley et al. (25) that these measurements are affected by the post-weaning age rather than the feeding of organic acids.

To recapitulate the results of the present experiment, it can be stated that medium-chain fatty acids contained in the Selacid feed preparation can improve piglets' health and performance.

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