

## **Fatty acid composition of porcine while adding amino acid, vitamin mineral and oily components under the conditions of industrial pollution**

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### **Abstract**

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The work gives the data on qualitative composition of saturated, monounsaturated and polyunsaturated fatty acids, as well as on their proportion in liver, longest back, iliac and thigh muscles of pigs bred under the conditions of industrial pollution. Also, the work gives the data on changes in fatty acid profile of porcine liver and muscles after 14 days of the use of two different biologically active complexes. The article describes both positive and negative effects of the use of biologically active complexes and their influence on fatty acid profile of liver and muscles of pigs bred under the conditions of environmental industrial pollution and given protein-imbalanced diet. The research is executed at the expense of a grant of the Russian scientific fund (project No. 16-16-00071).

*Keywords:* pigs; industrial pollution; fatty acids; muscles; iliac muscle

### **Introduction**

The Ural area is the territory where plants, animals (including agricultural ones), as well as humans, are subject to the influence of unfavorable ecological factors, including man-made genesis. Industrial pollution, which is the source of various toxicants, including heavy metals, takes a special place among the factors of negative influence.

Also, heavy metals have a negative effect on the organisms of animals and humans, that is inactivate and denature influence of heavy metals on ferments and other proteins that results in metabolic disorder, as well as in reduced antioxidant functions of animals and humans.

One of the links of antioxidant system is monounsaturat-

ed and polyunsaturated fatty acids, which protect cell membranes from free radicals, because of double bonds available in their structure.

Industrial pollution facilitates increased free-radical process in porcine organism, that leads to reduced content of polyunsaturated fatty acids, as well as to malfunction of the organs, resulted in worse quality of the products.

The most important essential fatty acids are linolic ( $\omega$ -6) and linolenic ( $\omega$ -3) fatty acids, as they are precursors of a number of regulatory molecules in organisms of animals and humans, namely icosanoids (Lands, 1992; Mohrhauer & Holman, 1963). So, it is important to manufacture animals' products with balanced content of polyunsaturated fatty acids, especially under the conditions of industrial pollution.

## Materials and Methods

For the experiment on formation of full-valued proteogenic composition of tissues of parenchymal porcine organs, three groups of animals kept under the conditions of industrial pollution were created. Every group had 25 animals with average weight of 60 kg at the moment of the start of the experiment. Together with the feedstuff used at the farm under the experiment, the pigs from the experimental group #1 were given biologically active complex №1, including: cholecalciferol, pyridoxin, niacin, vitamin C, magnesium citrate, complex of Calcium with methionine hydroxy analog (MHA), and lysine. The animals from the from the experimental group #2 were given the complex including: isolate of soya, pyridoxin, niacin, vitamin C, complex of Calcium with methionine hydroxy analog (MHA), lysine, threonine, valine and tryptophane, linseed, olive and sesame seed oils, and leaves of *Urticadioica*. The pigs from the control group were given only standard feedstuff. The duration of the use of the complex was 14 days.

After the end of the experiment the pigs were killed at a special slaughterhouse. After slaughter samples of muscle tissue (longest back muscle (*Musculus longissimus*), iliac muscle (*Musculus longissimus*), and thigh muscle) and liver were taken from every animal slaughtered.

Determination of fatty acid content of porcine muscles and liver was done according to GOST P 55483–2013. Fatty acid methyl esters were prepared according to GOST 31665–2012. Separation and registration of the components of the prepared sample were done using a gas chromatograph Trace GC Ultra with mass-spectrometer DSQII (Thermo Scientific, USA). The

conditions of the analysis: carrier gas – helium; sample input mode – without split ratio; volume of injection – 1  $\mu$ l; temperature of an injector – 230°C; column – capillary TR-5MS 25 m x 0.25 mm x 0.25  $\mu$ l; volume rate of carrier gas through column - 1 ml/min; temperature profile of column heating oven – 70°C (5 min), 5°C/min up to 130°C, 2°C/min up to 180°C, 5°C/min up to 240°C, 240°C (11 min), 30°C/min up to 280°C, 280°C (10 min); temperature of ion source – 200°C.

Identification of the components of s sample was done using the data on holding time of fatty acid methyl esters of standard mix (F.A.M.E. Mix C8-C24, Supelco), which was analyzed under the conditions similar to the ones for samples. For methyl esters, which were absent in standard mix, identification was done by means of comparing experimental mass-spectrum with partitioned data (Wiley9, NIST11, LIBRARY FAMES).

The results obtained in the course of the research were analyzed using the method of mathematical statistics, with averaging and standard deviation.

Normality of sampling was determined by means of criteria Shapiro-Wilk.

To evaluate availability of verified difference between the groups according to various parameters in case of normal distribution of samples – method ANOVA, in case of non-compliance with the conditions – criteria Mann-Whitney.

## Results and Discussions

The analysis of total content of saturated fatty acids in liver of the pigs given biologically active complex №1, did not show any verified difference as compared to the control

**Table 1. Proportion of saturated fatty acids in liver**

Liver	Saturated												
	Caprylic C8:0	Caprinic C10:0	Lauric C12:0	Tridecyl C13:0	Myristic C14:0	Pentadecyic C15:0	Palmitic C16:0	Margaric C17:0	Stearic C18:0	Arachic C20:0	Behenic C22:0	Lignoceric C24:0	Total of saturated fatty acids
Control	0.00	0.00	0.07	0.05	0.36	0.08	19.64	1.71	35.61	0.12	0.43	0.76	58.83
Std.Dev.	0.00	0.00	0.02	0.05	0.07	0.01	3.75	0.46	4.66	0.02	0.05	0.46	1.22
Experimental 1	0.00	0.00	0.21	0.09	0.49	0.21	19.35	1.96	31.46	0.19	0.96	3.30	58.23
Std.Dev.	0.00	0.00	0.07	0.09	0.06	0.17	1.42	0.34	5.42	0.11	0.42	2.66	1.32
±% (Control/Exp1)	0.0	0.0	181.8	75.0	38.3	152.0	-1.4	14.8	-11.7	65.7	122.5	332.8	-1.0
p-value	1.000	1.000	0.081	0.513	0.081	0.081	0.663	1.000	0.383	0.663	0.081	0.081	0.663
Experimental 2	0.00	0.00	0.00	0.02	0.21	0.06	15.69	1.31	29.07	0.05	0.37	21.20	67.97
Std.Dev.	0.00	0.00	0.00	0.02	0.04	0.02	0.72	0.06	0.49	0.02	0.02	0.71	1.59
±% (Control/Exp2)	0.0	0.0	-100.0	-56.3	-41.1	-28.0	-20.1	-23.4	-18.4	-57.1	-14.7	2676.9	15.5
p-value	1.000	1.000	0.081	0.663	0.081	0.190	0.383	0.663	0.081	0.081	0.127	0.081	0.081

group. However, there was a tendency to changes in proportion of saturated fatty acids, namely increased content of lignoceric (C24:0) fatty acid by 4,3 times, as well as of a number of other fatty acids (Table 1).

At the same time, total content of saturated fatty acids in liver of the pigs from the second experimental group was 15,5% higher than in the control group. Increased content of fatty acids is caused by significant increase in content of lignoceric fatty acid and decreased content of other saturated fatty acids (Scorletti & Byrne, 2013).

In liver of the experimental group №1 no significant changes in content of monounsaturated and polyunsaturated

fatty acids were stated. However, content of palmitoleic acid went up, that spoke for its increased endogenous synthesis (Table 2).

In liver of the experimental group №2 reduced content of monounsaturated and polyunsaturated fatty acids was stated that proves activation of aerobic metabolic process in liver and the use of fatty acids as antioxidants, and probably for synthesis of eicosanoids.

In longest back muscle of pigs from the experimental group №1 no changes in total content of fatty acids were stated, whereas in the experimental group №2 their verified reduced content by 4,5% was stated, as compared with the

**Table 2. Proportion of mono- and polyunsaturated fatty acids in porcine liver**

Liver	Monounsaturated							Polyunsaturated		
	Stearic C10:1	Myristoleic C14:1	Palmitoleic C16:1	Oleic C18:1	Gondoinic C20:1	Erucic C22:1	Total of monoun-saturated fatty acids	Linoleic C18:2	Linolenic C18:3	Total of polyun-saturated fatty acids
Control	0.00	0.18	1.26	22.06	0.10	0.12	23.72	17.11	0.34	17.45
Std.Dev.	0.00	0.07	0.12	2.01	0.03	0.05	2.16	1.21	0.11	1.28
Experimental 1	0.00	0.27	1.67	22.72	0.09	0.08	24.82	16.57	0.38	16.95
Std.Dev.	0.00	0.07	0.32	1.98	0.02	0.07	2.22	2.68	0.03	2.71
±% (Control/Exp1)	0.0	48.1	32.6	3.0	-10.0	-34.3	4.7	-3.2	12.9	-2.8
p-value	0.827	0.383	0.081	0.663	0.663	1.000	0.663	0.663	0.663	0.663
Experimental 2	0.00	0.14	0.89	15.43	0.28	0.88	17.62	14.17	0.24	14.41
Std.Dev.	0.00	0.01	0.09	0.81	0.04	0.13	1.05	0.61	0.07	0.55
±% (Control/Exp2)	0.0	-20.4	-29.2	-30.1	180.0	654.3	-25.7	-17.2	-27.7	-17.4
p-value	0.827	0.663	0.081	0.081	0.081	0.081	0.081	0.081	0.383	0.081

**Table 3. Proportion of saturated fatty acids in longest back muscle**

Longest back muscle	Saturated												
	Caprylic C8:0	Caprimic C10:0	Lauric C12:0	Tridecyl C13:0	Myristic C14:0	Pentadecyic C15:0	Palmitic C16:0	Margaric C17:0	Stearic C18:0	Arachic C20:0	Behenic C22:0	Lignoceric C24:0	Total of saturated fatty acids
Control	0.02	0.10	0.08	0.02	1.18	0.06	24.82	0.16	10.21	0.07	0.96	0.69	38.38
Std.Dev.	0.00	0.01	0.01	0.03	0.06	0.06	0.66	0.01	0.76	0.02	0.13	0.92	1.06
Experimental 1	0.02	0.11	0.10	0.06	1.17	0.04	24.70	0.17	10.81	0.09	0.91	0.66	38.84
Std.Dev.	0.01	0.00	0.02	0.03	0.11	0.02	0.96	0.01	1.60	0.04	0.26	0.22	2.14
±% (Control/Exp1)	-16.7	6.5	26.1	142.9	-1.4	-33.3	-0.5	2.0	5.9	22.7	-5.2	-3.9	1.2
p-value	0.663	0.275	0.190	0.275	1.000	0.827	1.000	1.000	0.663	0.827	0.663	0.663	1.000
Experimental 2	0.02	0.18	0.07	0.05	1.04	0.06	23.75	0.15	10.06	0.17	0.11	0.99	36.66
Std.Dev.	0.01	0.07	0.01	0.06	0.10	0.04	0.83	0.04	1.34	0.06	0.03	0.75	0.39
±% (Control/Exp2)	-16.7	77.4	-8.7	114.3	-12.1	0.0	-4.3	-8.2	-1.4	136.4	-88.6	43.5	-4.5
p-value	0.663	0.081	0.663	0.827	0.081	0.827	0.275	0.663	0.827	0.081	0.081	0.383	0.081

control group, towards the optimum proportion (optimum content of monounsaturated fatty acids - 30%) (Table 3).

Changes in percentage of monounsaturated fatty acids in longest back muscle of the pigs from the experimental groups were not verified. However, in longest back muscle of the pigs from the experimental group №2 content of erucic acid increased by 25 times (Table 4).

Content of polyunsaturated fatty acids in longest back muscle of the pigs from the first experimental group did not

show any significant changes, but there was a tendency to reduction of their content in general. At the same time, content of polyunsaturated fatty acids in the muscle of the second experimental group showed tendency to increase, towards the optimum figures and was  $7.02 \pm 6.7\%$  (optimum content of polyunsaturated fatty acids – 10%) (Bibus & Lands, 2015).

In iliac muscle of the pigs from experimental groups №1 and №2 no changes in total content of saturated fatty acids were stated (Table 5).

**Table 4. Proportion of mono- and polyunsaturated fatty acids in longest back muscle**

Longest back muscle	Monounsaturated							Polyunsaturated		
	Stearic C10:1	Myristoleic C14:1	Palmitoleic C16:1	Oleic C18:1	Gondoinic C20:1	Erucic C22:1	Total of monoun-saturated fatty acids	Linoleic C18:2	Linolenic C18:3	Total of polyunsaturated fatty acids
Control	0.00	0.03	4.04	53.78	0.27	0.01	58.13	3.33	0.16	3.49
Std.Dev.	0.00	0.01	0.32	0.63	0.03	0.01	0.97	0.46	0.01	0.46
Experimental 1	0.00	0.04	3.97	54.01	0.24	0.01	58.27	2.74	0.14	2.89
Std.Dev.	0.00	0.01	0.54	1.81	0.03	0.01	2.02	0.49	0.03	0.53
±% (Control/Exp1)	0.0	10.0	-1.7	0.4	-11.0	0.0	0.2	-17.5	-12.2	-17.3
p-value	0.827	0.663	0.663	1.000	0.275	0.827	1.000	0.383	0.663	0.383
Experimental 2	0.00	0.03	3.42	51.96	0.74	0.17	56.32	4.40	2.62	7.02
Std.Dev.	0.00	0.01	0.22	6.41	0.27	0.13	6.35	2.38	4.36	6.69
±% (Control/Exp2)	0.0	-10.0	-15.5	-3.4	169.5	2500.0	-3.1	32.3	1502.0	101.1
p-value	0.827	0.827	0.081	0.663	0.081	0.081	1.000	1.000	0.663	1.000

**Table 5. Proportion of saturated fatty acids in iliac muscle**

Iliac muscle	Saturated												
	Caprylic C8:0	Caprinic C10:0	Lauric C12:0	Tridecyl C13:0	Myristic C14:0	Pentadecylic C15:0	Palmitic C16:0	Margaric C17:0	Stearic C18:0	Arachic C20:0	Behenic C22:0	Lignoceric C24:0	Total of saturated fatty acids
Control	0.02	0.10	0.09	0.06	1.15	0.07	25.13	0.28	11.30	0.10	0.81	0.73	39.84
Std.Dev.	0.01	0.01	0.02	0.05	0.07	0.02	0.51	0.03	0.68	0.05	0.07	0.19	0.71
Experimental 1	0.02	0.10	0.13	0.03	1.21	0.05	25.43	0.31	12.75	0.08	0.76	0.70	41.56
Std.Dev.	0.00	0.01	0.03	0.03	0.19	0.02	1.16	0.04	1.43	0.01	0.06	0.42	2.44
±% (Control/Exp1)	20.0	3.3	46.2	-55.6	4.9	-33.3	1.2	9.5	12.8	-19.4	-6.2	-3.7	4.3
p-value	0.663	0.827	0.275	0.513	0.827	0.383	0.663	0.383	0.190	1.000	0.383	0.663	0.663
Experimental 2	0.01	0.16	0.08	0.04	1.05	0.07	24.58	0.24	12.65	0.17	0.19	1.25	40.48
Std.Dev.	0.00	0.01	0.01	0.03	0.07	0.05	0.56	0.08	0.37	0.01	0.02	0.41	1.37
±% (Control/Exp2)	-40.0	60.0	-7.7	-38.9	-9.0	0.0	-2.2	-13.1	11.9	61.3	-76.1	71.2	1.6
p-value	0.275	0.081	0.663	0.663	0.190	1.000	0.383	0.663	0.081	0.127	0.081	0.190	0.827

In iliac muscle of experimental groups №1 and №2 no significant changes in total content of monounsaturated fatty acids was stated. However, in the muscle of the second group significant increase in content of gondoinic and erucic fatty acids was stated (Table 6).

Content of polyunsaturated fatty acids in iliac muscle of the pigs from the first experimental groups had a tendency to decrease, whereas in the second group there was a tendency towards increase in content of polyunsaturated fatty acids.

In thigh muscle of pigs from the first experimental group total content of saturated fatty acids was not significantly different from the one in the control group, but there is a tendency towards decrease in total content of saturated fatty acids. Also, there is a strong tendency towards increase in content of caprilic, caprinic, tridecyl and arachic fatty acids. In thigh muscle of the second experimental group there is a tendency towards decrease in total content of saturated fatty acids, but at the same time there is a string tendency towards

**Table 6. Proportion of mono- and polyunsaturated fatty acids in iliac muscle**

Iliac muscle	Monounsaturated							Polyunsaturated		
	Stearic C10:1	Myristoleic C14:1	Palmitoleic C16:1	Oleic C18:1	Gondoinic C20:1	Erucic C22:1	Total of monounsaturated fatty acids	Linoleic C18:2	Linolenic C18:3	Total of polyunsaturated fatty acids
Control	0.00	0.06	3.68	49.69	0.16	0.00	53.59	6.30	0.27	6.56
Std.Dev.	0.01	0.01	0.43	0.98	0.14	0.01	1.54	1.39	0.04	1.41
Experimental 1	0.00	0.07	3.43	48.78	0.20	0.00	52.49	5.68	0.28	5.96
Std.Dev.	0.00	0.01	0.31	3.03	0.04	0.01	3.03	0.59	0.02	0.61
±% (Control/Exp1)	0.0	23.5	-6.7	-1.8	25.0	0.0	-2.1	-9.8	6.3	-9.1
p-value	0.663	0.275	0.383	0.663	0.827	0.827	0.663	0.663	0.827	0.663
Experimental 2	0.00	0.05	3.17	48.43	0.69	0.16	52.50	6.76	0.26	7.01
Std.Dev.	0.00	0.02	0.12	3.55	0.09	0.06	3.54	2.19	0.11	2.30
±% (Control/Exp2)	-100.0	-11.8	-13.8	-2.5	333.3	4700.0	-2.0	7.3	-3.8	6.9
p-value	0.663	0.513	0.383	1.000	0.081	0.081	1.000	1.000	1.000	1.000

**Table 7. Proportion of saturated fatty acids in thigh muscle**

Thigh muscle	Saturated												
	Caprilic C8:0	Caprinic C10:0	Lauric C12:0	Tridecyl C13:0	Myristic C14:0	Pentadecynic C15:0	Palmitic C16:0	Margaric C17:0	Stearic C18:0	Arachic C20:0	Behenic C22:0	Lignoceric C24:0	Total of saturated fatty acids
Control	0.01	0.08	0.10	0.03	1.00	0.08	23.50	0.23	10.09	0.05	0.98	0.86	37.01
Std.Dev.	0.00	0.01	0.02	0.03	0.07	0.04	0.54	0.08	0.69	0.06	0.06	0.39	1.33
Experimental 1	0.02	0.12	0.14	0.09	1.09	0.06	23.57	0.22	10.26	0.13	0.91	1.06	37.68
Std.Dev.	0.01	0.02	0.10	0.06	0.10	0.03	0.31	0.03	0.89	0.03	0.15	0.65	1.09
±% (Control/Exp1)	133.3	50.0	48.3	250.0	8.7	-20.8	0.3	-5.7	1.7	143.8	-6.8	22.8	1.8
p-value	0.081	0.081	0.827	0.190	0.190	0.827	1.000	1.000	1.000	0.190	0.663	0.663	0.663
Experimental 2	0.01	0.18	0.08	0.03	0.98	0.05	22.28	0.19	9.70	0.17	0.17	1.37	35.22
Std.Dev.	0.01	0.04	0.03	0.01	0.17	0.02	1.29	0.05	0.37	0.01	0.03	0.37	1.99
±% (Control/Exp2)	33.3	120.8	-17.2	0.0	-2.0	-41.7	-5.2	-20.0	-3.8	225.0	-82.3	59.1	-4.9
p-value	0.275	0.081	0.190	1.000	0.190	0.827	0.081	0.081	0.081	0.081	0.081	0.190	0.081

**Table 8. Proportion of mono- and polynonsaturated fatty acids in thigh muscle**

Thigh muscle	Monounsaturated							Polynonsaturated		
	Stearic C10:1	Myristoleic C14:1	Palmitoleic C16:1	Oleic C18:1	Gondoinic C20:1	Erucic C22:1	Total of monounsaturated fatty acid	Linoleic C18:2	Linolenic C18:3	Total of polynonsaturated fatty acids
Control	0.00	0.05	3.63	53.18	0.24	0.12	57.21	5.55	0.21	5.76
Std.Dev.	0.00	0.02	0.39	2.42	0.00	0.20	2.66	1.85	0.04	1.81
Experimental 1	0.00	0.05	3.91	53.92	0.20	0.00	58.08	4.05	0.20	4.25
Std.Dev.	0.01	0.01	0.34	1.62	0.02	0.00	1.36	0.63	0.02	0.64
±% (Control/Exp1)	0.0	7.1	7.8	1.4	-15.3	-100.0	1.5	-27.1	-3.2	-26.3
p-value	0.663	0.827	0.383	1.000	0.081	0.663	1.000	0.383	1.000	0.383
Experimental 2	0.00	0.05	3.36	54.07	0.80	0.22	58.50	6.07	0.20	6.27
Std.Dev.	0.00	0.02	0.40	4.00	0.09	0.08	3.59	1.55	0.05	1.60
±% (Control/Exp2)	0.0	14.3	-7.3	1.7	231.9	88.6	2.2	9.3	-3.2	8.8
p-value	0.827	0.513	1.000	0.190	0.081	0.081	0.190	1.000	0.127	1.000

increase in content of caprylic, caprinic, tridecyl and arachic fatty acids. In both cases a tendency towards decrease in content of saturated fatty acids is caused by increased use of those fatty acids as energy substrate for aerobic stages of ventilation (Table 7).

Total content of monounsaturated fatty acids in thigh muscle of the first and second groups does not have any verified differences as compared to the control group. At the same time, in thigh of the second group increases in content of gondoinic and erucic fatty acids was stated (Table 8).

Total content of polynonsaturated fatty acids in thigh muscle of the first experimental group had a tendency to decrease that is probably caused by depletion of its pool in that muscle. Content of polynonsaturated fatty acids in thigh muscle of the second experimental group did not undergo any significant changes. However, a tendency towards increase of their content as compared to the control group was stated.

## Conclusion

The results of the research done and analysis of the obtained data on qualitative composition and percentage ratio of saturated, monounsaturated and polyunsaturated fatty acids in liver and muscles of pigs bred under the conditions of industrial pollution and inadequate diet and given biologically active complexes, make it possible to speak about positive effect of biologically active complex №2 on fatty acid profile of liver and muscles of the pigs from the second group. The

effect was proved by increased content of polynonsaturated fatty acids and reduced percentage ratio of saturated fatty acids in muscles, as well as by the tendency towards normalization of content of polynonsaturated fatty acids in liver of pigs from that group.

At the same time, fatty acid profile of liver and muscles of the experimental group №1 did not show any significant changes, but there was a tendency towards decrease in content of polynonsaturated fatty acids, that speaks for inadequate diet of pigs bred under the conditions of industrial pollution, resulted in their increased catabolism when using biologically active complex №1.

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