Parameters of protein, lipid and mineral metabolism in pigs receiving a sorbent additive

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Abstract. This study provides evaluation of physiological status of sows and piglets during the period of raising and fattening and describes estimated concentrations, distribution and elimination of heavy metals and other toxicants from the animal body with a sorbent food additive introduced to their diet. We have studied the physiological status of sows and piglets they produced, under the effect of a novel sorbent additive made of raw hydroalumosilicates, and carried out physiological and biochemical assessment of prospects and reasonability of utilization of this additive in pig farming. This preparation in a dose of 120-150 mg/kg of the body mass was introduced to diet of pregnant sows, piglets at the phase of weaning and fattening pigs. Findings of our study allowed us to conclude that the investigated sorbent food additive had no negative effect on adequacy of vitamin levels in pigs in different age and gender groups. Increase in concentrations of some vitamins was attributable to reduction in the general toxic load on the animal body. At the same time, high ascorbic acid concentrations in blood of pregnant pigs and piglets not only activated different biological processes in the body but also facilitated removal of iron from ferritin. Thus, utilization of the sorbent food additive is a promising method for obtaining organic products.

1. Introduction
At the present moment environmental pollution with toxicants has become a global challenge. As we can hardly expect any positive changes in the foreseeable future, the search for efficient means of protection of farmed animals and animal products against industrial pollutants is a major line of research today. Use of food additives with absorbing, ion-exchanging and biologically active properties is a viable solution to this issue. In particular, highly oxidized cellulose, lithium compounds, bentonite, kaolinite, various zeolites, ascorbic acid and its derivatives and other preparations have been comprehensively tested for this purpose [1-3].

Sorbent additives fed to piglets have shown positive effects on their growth rate and normalized metabolic processes in the animal body. At the same time, they do not cause any dystrophic changes in the structure of epithelial cells in the gastrointestinal tract, liver, pancreas and kidneys [4-6].
2. Object and methods
In order to solve the problems posed, we carried out trials to investigate effects of the sorbent additive on physiological status and productivity of pregnant sows and their piglets in the phase of raising and fattening in the setting of a pig factory farm. In order to carry out a scientific and practical experiment aimed at investigation of the effects of the sorbent additive, we formed two animal groups (1st control group and 2nd experimental group) based on the analogy principle – each group included 50 animals. Pregnant sows were enrolled 40 days before farrowing. All the animals in the experiment were on the diet adopted by the farm, which was well-balanced by basic nutritional and biologically active substances. Pregnant sows in the second (experimental) group received the sorbent additive in a dose of 120 mg·kg⁻¹ of the body mass with their standard food. The sows were receiving the additive until farrowing and for five days after. During this period we evaluated physiological status of the animals. Piglets produced by sows in the second group were fed with 120 mg·kg⁻¹ of the additive from the weaning phase until the age of nine months. Thus, the experiment lasted for 300 days in total. The pigs were slaughtered when they were three and nine months old (as soon as the fattening phase was completed), and then samples of tissues and organs were taken and analysed in terms of their morphology and biochemistry.

We investigated effects of the sorbent additive on metabolism processes taking place in the animal body. Protein, lipid and mineral metabolism statuses were assessed by biochemical parameters of tissues and organs of pigs in different age groups.

3. Results
Examination of tissues and organs of three-month-old piglets showed that the sorbent additive in their diet caused no changes in levels of total lipids and protein in muscles and the liver. At the same time, cholesterol concentrations in meat tended to decrease (by 16.8%) and significantly increased in the liver (by 27%) (table 1).

| Parameters          | I – control | II – experiment | I – control | II – experiment |
|---------------------|-------------|-----------------|-------------|-----------------|
| crude protein, %    | 24.8±0.31   | 24.1±0.19       | 17.5±0.49   | 18.3±0.46       |
| protein, %          | 24.3±0.27   | 23.6±0.18       | 17.2±0.49   | 17.9±0.46       |
| lipids, %           | 2.44±0.048  | 2.71±0.197      | 3.99±0.573  | 3.34±0.364      |
| nitrates, mg/kg     | 24.9±0.37   | 22.4±2.03       | 33.3±2.48   | 26.3±1.13       |
| vitamin B₁, μg/g    | 30.7±1.82   | 32.7±0.53       | not analyze | not analyze     |
| vitamin B₂, μg/g    | 3.3±0.14    | 3.6±0.28        | not analyze | not analyze     |
| vitamin A, μg/g     | 97.1±7.23   | 108.3±10.41     | 2.51±0.095  | 3.05±0.155 a    |
| cholesterol, mg%    | 610.0±36.8  | 775.0±6.6 a     | 252.5±16.33 | 210.1±10.61     |

*p<0.05 (Mann-Whitney U-test).

The increase (p<0.05) in cholesterol concentrations in the liver is a rather natural process, since this organ is the place where cholesterol is produced and transformed.

The good news was that thiamine and riboflavin concentrations in the liver remained unchanged, and that nitrate levels in the liver and muscle tissue tended to decrease by 10.4% and 21.1%, respectively.

Vitamin A level is an essential parameter in evaluation of metabolism status. Introduction of the sorbent additive to formulated feed promoted significant increase (by 21.5%) in retinol concentrations in muscles, and its levels in the liver and spleen also tended to increase (by 11.5% and 37.6%, respectively).

We established close polynomial relationship between copper and cadmium salts concentrations in muscles and vitamin A levels. At the same time, higher vitamin A levels contribute to more profound involvement of zinc, iron and copper in metabolic processes.
The parameters estimated in the process of examination of tubular bones are noteworthy. Such bones of the piglets that received the sorbent showed increase in calcium and phosphorus concentrations by 21.6% and 31.4%, respectively, and at the same time iron, zinc, copper, cadmium and lead levels in the bones were low. The intensified calcium phosphate mineralization should be deemed positive. The mechanism of this phenomenon, most likely, includes replacement of some heavy metals with calcium and phosphorus. Reduced absorption of copper, cadmium and especially lead (which, at it is well-known, is an element deposited in bone tissue), resulted in mobilization of those elements from bones and concurrent intensification of calcium and phosphorus assimilation and increase in strength of bones of the piglets.

On the other hand, the sorbent additive contains up to 26 mass % of calcium oxide. Some of it was absorbed and penetrated into the circulatory system and, thus, reduced the amount of calcium withdrawn from bones to support other physiological systems of the animal body.

Significant correlations were found between heavy metals concentrations and calcium and phosphate salt levels in bones (table 2).

**Table 2.** Correlations between the content of heavy metals and calcium and phosphorus salts in the bone tissue of three-month-old piglets.

| Correlated indicator | Correlation coefficient | Regression equation |
|----------------------|------------------------|---------------------|
| Cu$^{2+}$ – Ca$^{2+}$ | -0.887411              | Y=6.295809326 – 41.67399532X+118.9263394X$^2$ |
| Cu$^{2+}$ – P$^{3+}$ | -0.859599              | Y=37.56885315 – 226.2825344X+384.5006800X$^2$ |
| Cd$^{2+}$ – Ca$^{2+}$ | -0.808744              | Y=34.45406422 – 697.8767424X+4528.194224X$^2$ |
| Cd$^{2+}$ – P$^{3+}$ | -0.813467              | Y=22.75443973 – 464.3618343X+2835.109648X$^2$ |
| Pb$^{2+}$ – Ca$^{2+}$ | -0.934729              | Y=42.29707500 – 52.24103469X+19.77938179X$^2$ |
| Pb$^{2+}$ – P$^{3+}$ | -0.888618              | Y=33.77264742 – 45.28435186X+17.34933345X$^2$ |
| Ca$^{2+}$ – P$^{3+}$ | -0.807917              | Y=8.460953579 – 1.479090759X+0.122095411X$^2$ |

Apparently, the sorbent additive introduced to piglet diet triggers positive changes in the entire animal body. Elimination of heavy metals not only results in mitigation of toxic “hurt” to physiological systems but also normalizes metabolic processes. Instead of being involved in detoxification processes, retinol actively engages in enzymatic reactions and, thus, contributes to metabolism, which is especially important during first months of life since newborn piglets have just poor vitamin A reserves and are to replenish them first with colostrum’s and milk and then with food. Higher retinol depositions activate protective functions and boost body resistance to infections and other adverse environmental factors.

Intensified calcium and phosphate mineralization in piglets that receive the sorbent additive contributes to formation of strong bones and reduces the risk of injury of their extremities in the setting of intensive farming.

Analysis of mineral composition of tubular bones revealed absence of any significant difference in calcium and phosphorus levels between the control and experimental groups of fattening pigs. This finding can be attributable, in the first instance, to completion of ossification processes and, in the second instance, to less intense elimination of heavy metals, which we have already discussed.

Analysis of chemical composition of the liver and longissimus muscle of nine-month-old pigs identified significant increase in retinol concentrations by 16.2% and 25.2%, respectively.

Some authors report a similar effect of zeolites introduced to diet of other animals [7-11]. Causes and mechanisms of this phenomenon are analogous to those described above. At the same time, it can
also be related to reduction in concentrations of toxic compounds (heavy metals and nitrates) in the liver, muscle and blood.

When the sorbent additive was used in formulated feed, nitrate ion concentrations tended to reduce in the liver and muscle (by 16.9% and 27.7%, respectively).

It has been proved that nitrates and nitrites negatively affect carotene and vitamin A assimilation in the animal body.

In addition to possible effect of nitrates, in the process of investigation of vitamin A metabolism it must be reasonable to compare concentrations of retinol and its metabolites in the liver, blood and muscle tissue.

Analysis of the findings confirmed the negative effect of nitrates on the amount of retinol reserves in the liver of pigs and proved existence of close correlation between concentrations of this vitamin in the liver, blood and muscles.

It should be noted that lower concentrations of toxicants in blood facilitate normalization of vitamin A metabolism in the liver and muscles. Thus, findings of this study give evidence to unquestionable relationship between toxicant levels and retinol concentrations in organs and tissues and to positive effects of the sorbent additive on adequacy of vitamin levels in animals.

Regression equations (table 3) can be used to compute vitamin A levels in the liver and muscles by concentrations of basic xenobiotics in blood.

Table 3. Correlations between the content of toxic compounds and vitamin A in the liver, muscles and blood of fattening pigs.

| Correlated indicator | Correlation coefficient | Regression equation |
|----------------------|-------------------------|---------------------|
| blood – liver         |                         | Y=304.1002611 + 69.10417054X–73.58815959X² |
| NO₃⁻ – vitamin A      | -0.924006               | Y=266.7872718 + 7576.446207X–275353.3575X² |
| Cd²⁺ – vitamin A      | -0.865233               | Y=–993.4254389 + 20604.21998X–81003.76248X² |
| Pb²⁺ – vitamin A      | -0.801140               |                         |
| blood – muscles       |                         | Y=3.656597950 + 0.509046020X–0.911196911X² |
| NO₃⁻ – vitamin A      | -0.975133               | Y=–1.255054299 + 500.7531878X–12926.16508X² |
| Cd²⁺ – vitamin A      | -0.922335               | Y=–25.33267683 + 447.2708801X–1727.778653X² |
| Pb²⁺ – vitamin A      | -0.844406               |                         |

4. Conclusion

Further, the still unclear situation with cholesterol concentrations in the animal body is to be noted. We have already discussed cholesterol levels in the liver. However, we have failed to find a satisfactory explanation for this problem. No significant changes were detected in blood and muscles of pigs in different age groups. Nevertheless, in muscles of three-month-old piglets the levels tended to reduce, and in fattening pigs it tended to increase. The question of causation is yet to be answered. At the same time, it seems that the sorbent additive does have some effect on cholesterol synthesis and degradation.

Long-time intake of the additive results in changes in protein metabolism in pigs. Protein concentrations in muscles and protein quality parameters (tryptophan-to-oxyproline ratio) increased significantly by 7.6% and 8.5%, respectively. This finding gives evidence to activation of protein biosynthesis in tissue. At the same time, the protein synthesized becomes more wholesome and the process of toxicant neutralization requires less energy.
Higher protein levels are also related to decrease in cadmium load on the body since it is a well-known fact that cadmium impedes protein synthesis at the translation initiation stage by disrupting polysome formation, while the elongation process accelerates.

The increase in protein concentrations in muscle tissue also indirectly suggests that the sorbent additive has no negative effect on the amino acid pool in the animal body, which is maintained by processes of enzymatic protein hydrolysis in the digestive tract, absorption of amino acids in the small intestine, and cellular cathepsins-mediated proteolysis.

Ultimately, all the metabolism parameters investigated give evidence to absence of any metabolic disorders triggered by the sorbent additive added to animal food. Moreover, the findings suggest that intake of the additive activates protein, lipid and mineral metabolism in pigs with concurrent detoxification and, thus, results in improvement of physiological status and productivity of animals.

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