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## The influence of feed additive “Sylymevit” on indicators of the immune system of piglets at weaning

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In the development of adaptive reactions during weaning, biological mechanisms are involved with the complete mobilization of the functional reserve, an increase in the level of catecholamines, corticosteroids, and mediators, which is accompanied by a violation of the nitrogen balance in the body, enzymopathy, and endotoxemia—the work aimed to investigate humoral and non-specific immunity indicators in piglets after weaning. Experiments were conducted based on the Ltd “KOSHET” of the Mukachiv district of the Zakarpattia region. Two groups of piglets were formed – control and experimental in the number of 10 individuals in each group, selected according to the principle of analogs – age, breed, and body weight. Weaning piglets from sows at 28 days suppress the humoral link of natural resistance. A decrease in lysocin and bactericidal activity of blood serum was established at  $40.56 \pm 1.75$  and  $21.22 \pm 1.05$  %. The content of circulating immune complexes in the blood of piglets of the control group remained high after weaning. Along with a decrease in the activity of the humoral link of immunity in weaned piglets, suppression of the non-specific link of the immune system was established, manifested by a decrease in phagocytic activity and a decrease in the number of phagocytes. Feeding the feed supplement “Sylymevit” to the piglets of the research group helped to strengthen the immune status of the piglets' bodies. In piglets of the research group on the 35th day of life, when feeding the feed additive “Sylymevit”, an increase in the phagocytic activity of neutrophils by 4.26 % was established. Similar differences were obtained regarding the effect of the Butaselmavit-plus feed additive on the phagocytic number and the phagocytic index, in particular, in piglets of the experimental group on the 35th day of the experiment; they were 7.63 and 11.1 % higher than in control. These research results confirm the effectiveness of using milk thistle, methifen, selenium, and vitamins as part of the feed supplement “Sylymevit” for piglets before and after weaning to activate the protective systems of their body.

**Key words:** oxidative stress, immune system, piglets, spotted thistle, sylymevit.

### Introduction

The introduction of intensive technologies in pig farming leads to the early weaning of piglets from sows, which causes stress, disruption of metabolic homeostasis, and an increase in free radical processes in the body. During the weaning of piglets, non-specific changes occur, which lead to the depletion of antioxidant potential

and a decrease in immunobiological reactivity. This is due to a weak level of adaptation processes in the body and an immunodeficiency state, which leads to a high frequency of diseases and a high percentage of mortality (Ariza-Nieto et al., 2011; Krempa et al., 2021; Martyshuk et al., 2021).

In conditions of oxidative stress, there is an increase in the intensity of radical formation, which leads to an in-

crease in lipid peroxidation processes (Ahmad et al., 2011; Martyshuk et al., 2019). Peroxide oxidation at almost all stages of its course forms several active products that result from the interaction of free radicals both among themselves and with biological macromolecules (Chala & Rusak, 2016; Martyshuk et al., 2018; 2022). It is important to emphasize that the increased formation of primary free radicals is a side effect of the increased intensity of biochemical reactions in response to the action of the stress factor – weaning from the sow.

The essential biochemical mechanism that affects the reduction of resistance and the occurrence of oxidative stress at these moments of life is the sharp and long-term activation of free radical oxidation and the formation and accumulation of products of oxidative modification of lipids and proteins in the body (Cherkashina & Petrenko, 2006; Lavryshyn et al., 2016; Gutj et al., 2017; 2019; Martyshuk et al., 2020).

As a result of early weaning, stress reduces the growth intensity of piglets and the activity of bone marrow cells, the number of erythrocytes, and the level of thyroid hormones in the blood. In the body of piglets, due to the effects of stress, the mass of internal secretion glands increases, and morphological changes in their structure also occur. A vital role in the development of stress syndrome in piglets after weaning from the sow is played by hormones of the adrenal cortex - glucocorticoids, the level of which in the blood increases significantly since the suppression of immune processes in piglets against the background of the body's stress reaction is due to the immunotropic effect of glucocorticoids. They also contribute to strengthening the catabolism of proteins and fats, increasing the sugar content in the blood and glycogen in the liver, inhibiting the formation of antibodies, and disrupting cellular immune reactions (Bulter et al., 2006; Chen et al., 2018; Martyshuk & Hutyi, 2021).

In the development of adaptive reactions during weaning, biological mechanisms are involved with the complete mobilization of the functional reserve, an increase in the level of catecholamines, corticosteroids, and mediators, which is accompanied by a violation of the nitrogen balance in the body, enzymopathy, and endotoxemia (Khalak et al., 2019; Khalak & Gutj, 2020; Khalak et al., 2020; Vyslotska et al., 2021).

In recent years, practical and cost-effective new complex drugs have been successfully developed that help prevent the adverse effects of stress and provide the necessary conditions for care and maintenance (Martyshuk et al., 2016; Khariv et al., 2016; 2017). The use of natural substances is auspicious in this context (De Lange et al., 2010; Czech et al., 2018; Gutj et al., 2022). Literary sources contain reports on the stimulating effect of milk thistle, fat-soluble vitamins, selenium, and metiphen on the activity of animal immune and antioxidant systems. However, these studies are fragmentary, so a detailed study and generalization of this topic is necessary. A comprehensive analysis of the effect of these substances on the antioxidant potential and immune function of the animal body under conditions of oxidative stress is fundamental. Conducting such research is relevant, as it paves the way for developing scientifically based methods

of managing adaptive and protective processes in animals, particularly piglets, during weaning from sows.

### The aim of the research

The work aimed to study the influence of the feed additive “Sylimevit” on indicators of the humoral and non-specific links of the immune system.

### Material and methods

Experiments were conducted based on the Ltd “KO-SHET” of the Mukachiv district of the Zakarpattia region. Two groups of piglets were formed – control (C) and experimental (E) in the amount of 10 individuals in each group, selected according to the principle of analogs – age, breed, and body weight. During the suckling period, piglets were kept under the sow in special machines, had constant access to the mother, and free access to the concentrated feed from the age of 5 days. Animal feeding was carried out following the norms for a given age of pigs. Before conducting research, a clinical and physiological examination of piglets was carried out, and their general condition and activity when eating feed were considered. On the 28th day of life, the piglets were weaned from the sow and regrouped from different nests for further maintenance during the period of fattening and rearing with a change in the structure of the diet, which served as technological stress for the animals' bodies. Starting from the age of 5 days, piglets of all groups were fed pre-starter compound feed. From the age of 21 to 40 days, the piglets of the research group were additionally fed the feed additive “Sylimevit” at a dose of 100 mg/kg of body weight per day.

The research material was blood, which was collected in the morning before feeding the animals by puncture of the cranial vena cava on the 20th day of life (the period before weaning), on the 25th day of life (the period before weaning), on the 30th day of life (2 days after weaning), on 35 days of life (7 days after weaning), on 40 days of life (12 days after weaning).

Lysozyme activity of blood serum was determined using a daily culture of *Micrococcus lysodeicticus* strain VKM-109 as a test microbe by the nephelometric method; optical density was measured at a wavelength of 540 nm. Bactericidal activity in blood serum samples was studied according to this method by Yu. M. Markov (1968) using a daily culture of *E. coli* strain VKM-125. Photocolorimetry was performed before and after a 3-hour incubation (Vlizlo et al., 2012; Kotsumbas & Hryniv, 2016).

Determination of the content of circulating immune complexes in blood serum was carried out using a borate buffer. Selective precipitation of antigen-antibody complexes occurred under the influence of high molecular weight PEG with a mass of 6000 Da. The results were calculated by photocolorimetry of the density of the precipitate at a wavelength of 450 nm (Vlizlo et al., 2012).

The phagocytic reaction of blood neutrophils was assessed by phagocytic activity (PhA), phagocytic index (PhI), and phagocytic number (PhN) according to the method of V. S. Gostev (1950). Stabilized blood was incubated with a daily culture of *E. coli* strain VKM-125.

Smears were examined under a microscope in an immersion system. PhA was determined by the number of active neutrophils from 100 counted cells, PhI - by the number of phagocytosed microbial bodies by one active neutrophil, and PhN – by the number of phagocytosed microbial bodies per 100 counted neutrophils (Vlizlo et al., 2012).

The research results were analyzed using the Statistica 6.0 software package. Student's t-test assessed the probability of differences. The results were considered probable at  $P \leq 0.05$ .

### Results and discussion

The study of the humoral factors of the natural resistance of piglets showed that, concerning weaning at 20 days, the BABS value of animals of the control and experimental groups was 26.41 and 26.12 % (Table 1). After weaning, BABS decreased by 4.72 % in the blood of piglets of the control group compared to the indicators taken before weaning. At 35 and 45 days, the BABS in piglets of the control group remained at a low level, while in the experimental group of piglets, which were fed with the feed additive “Sylimevit”, this indicator was probably higher. Thus, in 30-day-old piglets of the experimental group, the BABS was higher by 3.82 %, and in 35-day-old piglets by 7.4 % compared to the control group of animals.

**Table 1**  
Bactericidal activity of blood serum of piglets under the action of feed additive “Sylimevit”, % ( $M \pm m$ ,  $n = 10$ )

Day of life	Group of piglets	
	Control	Experimental
20	26.41 ± 0.44	26.12 ± 0.48
25	31.27 ± 0.82	31.91 ± 1.10
30	21.69 ± 1.11	25.51 ± 0.72**
35	21.22 ± 1.05	28.62 ± 1.36**
40	22.10 ± 0.99	27.86 ± 1.04**

When researching the lysozyme activity of blood serum in piglets of the control and experimental groups, it was established that on the 20th day of the experiment, it was 40.51 % and 40.59 %. In 25-day-old piglets of the control group, LABS increased by 6.31 %, and of the experimental group – by 6.69 % (Table 2).

**Table 2**  
Lysozyme activity of the blood serum of piglets under the influence of the feed additive “Sylimevit”, % ( $M \pm m$ ,  $n = 10$ )

Day of life	Group of piglets	
	Control	Experimental
20	40.51 ± 0.71	40.59 ± 0.78
25	46.82 ± 0.80	47.28 ± 2.16
30	42.84 ± 0.92	49.51 ± 1.55**
35	40.56 ± 1.75	50.17 ± 2.00**
40	41.55 ± 2.02	49.72 ± 2.42*

As the results showed, weaning decreased the LABS in piglets of the control group on the 30th and 35th days of the experiment by 3.98 and 6.26 %. At the same time,

this indicator was probably higher in the experimental group where, on the 30th and 35-day-old piglets, it increased by 6.67 and 9.61 % compared to the indicators of the control group.

The content of CIC in the blood of piglets of the control group after weaning on the 30th day of the experiment was 14.7 % higher than in the period before weaning. Subsequently, the content of CIC in the blood of the control group of piglets decreased slightly but remained at a high level. When examining the level of CIC in the blood of the experimental group of piglets, which were given the feed additive “Sylimevit”, it was found that this indicator decreased by 7.9 and 9.3 % on the 30th and 35th days of the experiment, respectively, compared to the indicators of the control group of piglets (Table 3).

**Table 3**  
Circulating immune complexes in the blood of piglets under the influence of the feed additive “Sylimevit”, mmol/l ( $M \pm m$ ,  $n = 10$ )

Day of life	Group of piglets	
	Control	Experimental
20	71.26 ± 2.00	71.39 ± 2.06
25	72.36 ± 2.11	72.27 ± 1.81
30	81.71 ± 1.72	75.29 ± 2.25*
35	80.89 ± 2.14	73.33 ± 1.92*
40	80.45 ± 2.38	73.24 ± 2.41*

In general, the obtained data indicate the inhibitory effect of stress on indicators of natural resistance, especially humoral factors of protection in the piglets' body, and the effectiveness of the feed additive “Sylimevit” to normalize the detected disorders.

Along with a decrease in the activity of the humoral link of immunity in weaned piglets, suppression of the non-specific immune system was established, manifested by a decrease in phagocytic activity and a decrease in phagocytes.

The phagocytic activity of blood neutrophils in piglets before and after weaning reflects the ability of neutrophil granulocytes to phagocytize antigens foreign to the piglets' bodies. This function is ensured by the activity of blood opsonizing factors – antibodies and complement.

It was found that after weaning, the phagocytic activity of neutrophils decreased by 2.85 % compared to the initial values in piglets of the control group (Table 4). In the indicated period of the study, a slight increase in the phagocytic index was found in the blood of the control group, which was 7.49 units, respectively.

**Table 4**  
Phagocytic activity of piglet blood neutrophils under the influence of the feed additive “Sylimevit”, % ( $M \pm m$ ,  $n = 10$ )

Day of life	Group of piglets	
	Control	Experimental
20	45.41 ± 0.90	45.37 ± 0.59
25	45.65 ± 0.79	45.76 ± 0.68
30	42.80 ± 0.95	47.14 ± 1.02**
35	41.23 ± 0.87	48.86 ± 0.87***
40	48.57 ± 0.54	50.96 ± 0.95*

Feeding piglets with the feed supplement “Sylimevit” caused an activating effect on phagocytosis indicators (Table 5). Thus, on the 30th day of the experiment, the phagocytic activity of blood neutrophils in piglets of the experimental group was 4.34 % higher than in the control group.

**Table 5**

The phagocytic index of the blood of piglets under the influence of the feed additive “Sylimevit”, units (M ± m, n = 10)

Day of life	Group of piglets	
	Control	Experimental
20 day	7.33 ± 0.10	7.30 ± 0.11
25 day	7.41 ± 0.12	7.47 ± 0.10
30 day	7.49 ± 0.10	7.54 ± 0.08
35 day	7.64 ± 0.11	8.49 ± 0.11*
40 day	7.61 ± 0.19	8.10 ± 0.13*

Similar differences were obtained concerning the effect of the feed additive “Sylimevit” on PhN and PhI, in particular, in piglets of the experimental group on the 35th day of the experiment, they were 6.7 and 11.1 % higher than in control (Table 6).

**Table 6**

The phagocytic number of the blood of piglets under the action of the feed additive “Sylimevit”, units (M ± m, n = 10)

Day of life	Group of piglets	
	Control	Experimental
20 day	3.91 ± 0.13	3.98 ± 0.11
25 day	3.97 ± 0.10	4.10 ± 0.13
30 day	3.47 ± 0.11	3.91 ± 0.09**
35 day	4.05 ± 0.13	4.32 ± 0.10
40 day	4.11 ± 0.10	4.69 ± 0.12**

The above data indicate that the Sylimevit feed supplement's components stimulate phagocytosis, increasing the antimicrobial properties of blood cells.

These research results confirm the effectiveness of using milk thistle, methifen, selenium, and vitamins A, E, and C as part of the feed supplement “Sylimevit” for piglets in the period before and after weaning to activate the protective systems of their body.

### Conclusion

Weaning piglets from sows at 28 days suppress the humoral link of natural resistance. The use of the feed additive “Sylimevit” in piglets of the research group contributed to an increase in the bactericidal and lysozyme activity of blood serum, as well as an increase in the phagocytic activity of neutrophils and the phagocytic number during the weaning period.

### Conflict of interest

The authors declare that there is no conflict of interest.

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