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## **Research Article**

# **Evaluation of Immunity System in Newborns Pigs from Sow with Various Degrees of Immunological Load**

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### ABSTRACT

It was found that the level of nonspecific resistance of the organism and adaptive potential to environmental conditions are interdependent and interdependent. Therefore, the study of these mechanisms has the ultimate goal of justifying the intensity of the body's defense. Aim of this study is investigating the characteristics of the formation of immunobiological status in the offspring of the early postnatal period, depending on the immunological load of sows. Two groups of animals were divided; the first group is from a sow with intensive immunological load, the second is from a sow with low antigenic stimulation. Blood sampling was carried out before feeding, on 1, 5, 12 and 30 postnatal days. Analysis of peripherial immune cells shows the killer activity of T-lymphocytes and the production of interleukins are reduced in new borns compared to adults. In T-lymphocytes, 50-65% of a newborn are T-helpers carrying CD4 molecules, and 25-30% have a CD8 T-phenotype. T cells respond to infectious antigens (see Immunity and infection). However, the level of cytokine production is reduced. B-cell system of the newborn. High content of B-lymphocytes in the umbilical cord blood of the newborn is detected. level of B-lymphocytes carrying differentiating antigens CD19, CD22 in newborns is slightly higher than in adults. Piglets obtained from the second group up to one month of age had reduced quantitative and percentage indicators of immunobiological status in compared with piglets of the first group. The results of studies on the formation of the immune system of the body of newborn animals indicate that protective forces are a dynamic indicator and is determined by the immune specificity and antigenic load on the mother's body. This circumstance allows you to directionally influence the formation and manifestation of the body's defenses.

**Key words:** Immuno-pathological Mechanisms, Immuno-rehabilitation, Reactivity, Sensitivity, Pregnant animals, Newborn piglets

#### INTRODUCTION

Strengthening the immune system in newborns livestock is particularly important, and researchers are using a variety of strategies to this aim (Hlavová, 2014; Hussein, 2018). Increasing the survival and preservation of piglets in the early periods of ontogenesis is the most important of the tasks of veterinary medicine (Hlavová, 2014) . It is known that most of the departure of young animals occurs in the early postnatal period. This is due to several objective reasons, among which the primary role is played by the failure of the immune system (Brunse *et al.*, 2019; Milovanovic *et al.*, 2019).

It was found that the level of nonspecific resistance of the organism and adaptive potential to environmental conditions are interdependent and interdependent. Therefore, the study of these mechanisms has the ultimate goal of substantiating the intensity of the body's defense (Iraola *et al.*, 2016; Zheng *et al.*, 2018; Dennis *et al.*, 2019).

The technical result that has been achieved is reduced to the prevention of the immunodeficiency state of newborns in the fetal period by implementing an immunomodulating effect on the mother's body during the most stressful period (second half) of pregnancy (Hlavová, 2014).

It should be noted that the mother's body, in critical periods of pregnancy, is in a state of physiological immunodeficiency associated with intensive formation and development of the fetus, preparation of the body for the production of colostrum and milk. For sows, this period occurs in the second half of gestation (70-100 days) (Butler *et al.*, 2009).

At the same time, thanks to the principle of interconnectedness and interdependence of homologous functional systems in the mother-fetus-newborn complex,

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it is possible to activate the own biological reactions of the newborn by acting on the mother's body when the fetus is born to move to a more advanced level of functioning immediately after birth (Rørvang *et al.*, 2018).

It was established that newborns are born at earlier stages of morphofunctional development compared with other farm animals, in a state of physiological immunodeficiency, which necessitates the use of immunostimulating drugs precisely in the fetal period of development, thereby allowing receiving and growing more complete offspring (Socha-Banasiak *et al.*, 2017; Rørvang *et al.*, 2018).

The study aimed to study the characteristics of the formation of immunobiological status in the offspring of the early postnatal period, depending on the immunological load of sows.

We set the following research objectives:

- 1. To determine the formation of the immunobiological status of piglets in the early postnatal period.
- 2. To assess and the degree of physiological status in piglets.

#### MATERIALS AND METHODS

The experimental part was performed on pigs of large white breed. 2 groups of animals were formed, 10 animals each from the nest. The first group is from a sow with intensive immunological load, the second is from a sow with low antigenic stimulation. Blood sampling was carried out before feeding in the morning from the ear vein, on 1, 5, 12 and 30 days after birth. The following indicators were studied: the content of leukocytes and red blood cells in whole blood, the hemoglobin concentration and its level in one red blood cell, hematocrit, platelet count, platelet count, immunoglobulin concentration (A, G, M), the percentage of phagocytic activity of neutrophils (FAN%), phagocytic number (PS), phagocytic index (PI), bactericidal and lysozyme activity of blood serum. The indicators were determined by the following methods (Iraola et al., 2016; Zheng et al., 2018; Dennis et al., 2019):

- The content of hematological parameters on an automatic veterinary erythrohemometer PCE-90 VET.
- The morphological composition of the blood was studied by the quantitative content of red blood cells and leukocytes counted in the Goryaev's chamber and qualitative, counted in a smear stained according to Romanovsky-Giemsa (Kuhlmann, 2018).
- Concentration of immunoglobulins on a ChemWell automatic biochemical and enzyme immunoassay analyzer, version 5.1 (Revision E), using the Vector-Best sets Jg A, M, and G.
- Lysozyme activity of blood serum was determined by Novikov (1990).
- Bactericidal activity of blood serum according to Taylor (1983).
- Phagocytic activity of neutrophilic granulocytes, phagocytic index and phagocytic number according to Taylor (1983).
- Digital data were processed by biometric methods using applied computer programs Microsoft Excel and BioStat (2009).

#### **RESULTS AND DISCUSSION**

Given the data presented in Table 1, it can be observed that the main differences in the hematological examination of the blood of the piglets of the experimental groups were expressed in the trend of reduced indicators of the relative number of lymphocytes in volume.

A study of the functional activity of newborn lymphocytes showed that there is a marked reaction of blast transformation, both spontaneous and stimulated by antigens. However, there is a certain reactivity to bacterial antigens. The killer activity of T-lymphocytes and the production of interleukins are reduced compared to adults.

Among T-lymphocytes, 50-65% of a newborn are Thelpers carrying CD4 molecules, and 25-30% have a CD8 T-phenotype. T cells respond to infectious antigens (see Immunity and infection). However, the level of cytokine production is reduced.

B-cell system of the newborn. High content of B-lymphocytes in the umbilical cord blood of the newborn is detected. The number of B-lymphocytes with surface immunoglobulins in the first days of life is slightly reduced (Table 1).

B-lymphocytes with IgA receptors are usually not detected, on the contrary, lymphocytes with membrane IgD are detected in the amount of 13-14%, which is much higher than that in adults. Thus, newborns also have no deficiency of B-lymphocytes, but more of their immature subpopulations. The level of B-lymphocytes carrying differentiating antigens CD19, CD22 in newborns is slightly higher than in adults (Table 1).

No statistically significant differences between the groups were detected. However, if we talk about trends, piglets of the 1st group were characterized by higher values of these indicators, and piglets of the 2nd group were characterized by lower values.

Analyzing the data of the leukogram, increased content of lymphocytes is observed in piglets at the age of 7 days relative to the second group and lasts up to 30 days. Also in piglets of the 1st group, there is an increase in the number of segmented neutrophils (more mature cells) and monocytes due to a decrease in lymphocytes (Table 1).

The leukocyte count throughout the study was subject to fluctuations, characterized by a rise and fall with a general tendency to increase. If piglets of seven days of age had a white blood cell count of 4.19±0.57 in the second group and 6.22±0.13 in the first group, then 5.08±0.11 and 5.96±0 were already ten days old, 32, and in 30 days it was in the range of 11.05±0.12 and 13.17±1.1 x 109/L. The first group of animals exceeded their peers by 25.4%, 17.8% and 10.4% in the time aspect. A change in the ratio of segmented neutrophils to lymphocytes was also observed. If during the first seven days the relative number of the former was at the level of  $24.5\pm0.12 - 34.2\pm0.16\%$ , and the second - 35.1±0.24 - 40.7±0.18%. On the tenth day of life, these indicators became respectively 24.1±0.23 - 27.3±0.21 and 27.4±0.26 - 29.1±0.21%. On the thirtieth day, they had values of 34.8±3.3 - 47.1±2.6 and 35.3±6.3 - 36.9±7.8%. According to the content of individual forms of leukocytes, it was found that the quantitative content of segmented neutrophils is higher in the blood of animals of the first group, and in animals of the second group, this indicator is lower by 1.5–2.1%, which may be due to their higher activity.

**Table 1:** The content of T and B lymphocytes in peripheral blood depending on age (%).

Age	T lymphocytes		B lymphocytes		B-lymphocytes with surface immunoglobulins		
	1	2	1	2	G	М	А
	Group	Group	Group	Group			
1-2 day	48.01±0.26	32.05±0.15	32.26±0.26	23.58±0.14	13.05±0.15	2.91±0.77	-
5-6 day	49.04+0.28	44.63±0.11	26.18±0.18	24.47±0.24	14.63±0.17	2.59±0.45	-
12 <sup>th</sup> day	56.63±0.30	30.42±0.14	25.69±0.21	23.14±0.29	$10.42\pm0.14$	3.19±0.87	-
20 <sup>th</sup> day	60.82±0.32	44.71±0.17	25.05±0.24	24.17±0.15	9.01±0.15	4.54±0.46	-
1 month	60.44±0.30	51.81±0.19	24.44±0.17	25.82±0.11	7.81±0.16	5.66±0.11	3.25±0.17

Table 2: Serum Immunoglobulin Levels in Piglets.

Age	Concentration	in g / l			
	IgG	IgM	IgA	IgE, ME/l	
		1 Group			
1 day	$10.65 \pm 0.42$	0.16±0.02	$0.02 \pm 0.02$	0-10	
6 day	9.82±0.36	$0.19{\pm}0.03$	$0.08 \pm 0.04$	0-10	
12 day	9.78±0.34	$0.24{\pm}0.04$	$0.08 \pm 0.05$	0-20	
20 day	$8.94{\pm}0.28$	$0.27 \pm 0.03$	$0.12 \pm 0.04$	0-20	
1 month	10.11±0.35	$0.28 \pm 0.05$	$0.16 \pm 0.03$	0-20	
3 months	$5.02 \pm 0.12$	$0.21 \pm 0.16$	$0.32 \pm 0.12$	0-20	
4-6 months	$5.34 \pm 0.18$	0.36±0.18	$0.38 \pm 0.14$	0-30	
7-12 months	$7.55 \pm 0.22$	$0.76 \pm 0.27$	$0.54 \pm 0.16$	5-50	
		2 Group			
1 day	$8.21 \pm 0.12$	$0.12 \pm 0.02$	$0.01 {\pm} 0.02$	0-8	
6 day	$7.98 \pm 0.61$	$0.14 \pm 0.02$	$0.02 \pm 0.04$	0-9	
12 day	$8.97 \pm 0.42$	$0.24{\pm}0.04$	$0.01 {\pm} 0.05$	0-10	
20 day	8.19±0.15	$0.28 \pm 0.05$	$0.15 \pm 0.04$	0-10	
1 month	$9.04{\pm}0.75$	$0.20{\pm}0.08$	$0.16 \pm 0.03$	0-10	
3 months	4.19±0.12	0.20±0.10	$0.24{\pm}0.12$	0-10	
4-6 months	$4.92 \pm 0.18$	$0.34{\pm}0.11$	$0.31 \pm 0.14$	0-10	
7-12 months	$6.41 \pm 0.22$	$0.67 \pm 0.22$	$0.51 \pm 0.16$	5-40	

The progeny resistance indices reflect that the animals of the second group had a low level of cellular indices of protection, and the first group occupied a relatively higher position. They had higher rates of phagocytic (the phagocytic activity of neutrophils, phagocytic index, phagocytic number), lysozyme and bactericidal activity of blood serum.

During the entire period of research, the phagocytic ability of neutrophils was at the level of  $54.1\pm0.18$  -  $50.9\pm0.49\%$  in the first group and  $32.4\pm0.27$  - $42.5\pm0.39\%$  in the second and - for this, the absorbing and digesting activity of the first group were higher by 27, 15 and 38\%, respectively. Moreover, the lysozyme activity of blood serum was  $22.1\pm0.19$  -  $33.6\pm0.14\%$  and  $23.7\pm0.46$  -  $25.4\pm0.53\%$ , and the bactericidal activity was  $26.7\pm0.11$  -  $48.5\pm0.37\%$  and  $26.7\pm0.45$  -  $30.7\pm0.22\%$ .

The content of immunoglobulins (Ig) as shown (Table 2) in the blood serum of the first group from the seventh to the thirtieth day of life was wavy in nature as their own immune status developed, their number increased. Piglets obtained from sows of the second group up to one month of age had reduced quantitative indicators of immunobiological status in quantitative and percentage terms than piglets of the first farrow.

Decreasing in antigen with age on new born piglets in the table 1, is in agreement with Pasternak *et al.*, (2015), who stated that markedly decreases in cytokines (IL-10) in piglets after 72 hours postnatal. Also, this finding is in according to Nechvatalova *et al.* (2008), which show significant decreasing peripheral immune parameters in new born piglets continuously and immediately after born. high content of B-lymphocytes in the umbilical cord blood of the newborn is detected. And The number of B-lymphocytes with surface immunoglobulins in the first days of life is slightly reduced (Table 1), which this finding is in according to Nguyen *et al.* (2016) who reported a delay in development of systematic immunity in piglets, with measuring the immunoglobulins.

In present study, the level of B-lymphocytes carrying differentiating antigens CD19, CD22 in newborns is slightly higher than in adults (table. 1). This is may be due to down-regulation mechanism of B-cell-restricted molecule which it downregulates the signals between CD19 and B-cell receptor (Nitschke, 2005; Tedder *et al.*, 2005).

Whereas there is no statistically significant differences between the groups in B-lymphocytes carrying differentiating antigens CD19, CD22; piglets of the 1st group (high immune measures in mother) were characterized by higher values of these indicators, and piglets of the 2<sup>nd</sup> group (low immune measures in mother) were characterized by lower values. This observation is in agreement with Salmon *et al.* (2009), in their study on efficiency of humoral and cellular maternal immunity in piglets.

First group had higher rates of phagocytic, lysozyme and bactericidal activity of blood serum, may be due to high degree of maternal immune defense which is documented by Salmon *et al.*, (2009). Piglets obtained from sows of the second group (low immune measures in mother) up to one month of age had reduced quantitative indicators of immunobiological status (Table 2) in quantitative and percentage terms than piglets of the first farrow. So, the status of immune system in mother (sow) can be considered as immune heritage in piglets.

#### Conclusions

The formation of the immunity system in piglets is of a phase nature and depends on the system of immunity interventions in the form of vaccination of the mother's body. The results of studies on the formation of the immune system of the body of newborn animals indicate that protective forces are a dynamic indicator and is determined by the immune specificity and antigenic load on the mother's body. This circumstance allows you to directionally influence the formation and manifestation of the body's defenses. The study of the immunological reactivity of the organism of farm animals, thus, becomes relevant for understanding the pathogenesis of diseases, for rational pathogenetic therapy.

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