Morphofunctional characteristics of animals in the detection of embryonic antigens in the prenatal and postnatal periods

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Abstract. Diseases in newborn animals cause significant damage to animal husbandry. This is a complex problem, in which, along with such factors as the environment and the pathogen, an important role is played by the reaction of the body of newborns and their close connection with the mother's body. The study of enzyme relationships in the functional system «mother-fetus-newborn» can make a significant contribution to solving the problem of improving the safety of the population of newborn animals. Newborn animals have different degrees of functional maturity. Functional capacity of some organs and the system of the newborn, in comparison with the parent individuals, can be determined both genetically and by the conditions of intrauterine development. Currently, a sufficient number of facts have been accumulated that any deviations or violations of homeostasis parameters the mother's body affects the fetus and Vice versa. The main role in compensating for impaired functions belongs to the mother's body, but the fetus is also able to participate in these reactions to a certain extent. Functional integration of fetal and maternal homologous systems when performing homeostatic functions concerns the activity of the blood enzyme component. The aim of our research was to study quantitative and qualitative changes in the activity of blood enzymes in non-pregnant sows, in the first and second half of pregnancy and the postpartum period.

1 Introduction

The industrial technology of pork production makes corrective changes in the conditions of keeping animals associated with changes in the most active blood indicators [1, 3, 8, 16]. It becomes obvious that providing animals with nutrients alone is not a sufficient condition for maintaining the normal functional state of the animal organism, its high productivity and reproductive ability [4, 10, 12, 17].

As a result of the animal husbandry transition to an industrial basis and intensive rearing of productive animals, there is a need to establish and introduce new normative indicators of relative constancy in the internal environment of the animal organism [9, 13, 14, 15]..

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Numerous literature data indicate that the level of clearance of biologically active substances in the blood serum affects the fertilization, embryonic mortality and productivity of the animal [2, 4, 5].

Therefore, our studies of the activity of serum enzymes in non-pregnant and pregnant pigs are relevant at the moment [6, 7, 11].

The aim of our research was to study quantitative and qualitative changes in the activity of blood enzymes in non-pregnant sows, in the first and second half of pregnancy and the postpartum period.

2 Research materials and methods

Experimental studies were conducted on sows of a Large White breed on the farms in the Kurskoy district of the Stavropol territory. Histological studies were performed in the histological laboratory of the Scientific Diagnostic and Therapeutic Veterinary Center of the Stavropol State Agrarian University.

The research was carried out in the period 2018-2020 on the basis of the pig farm "Russia", located in the Novoaleksandrovsky district, Stavropol Territory. Biochemical parameters were determined on laboratory equipment in the "Scientific Diagnostic and Medical Veterinary Center" of the Stavropol State Agrarian University.

The blood of clinically healthy female pigs of Large White breed of the following groups was used as an object of research: non-pregnant, the first half of pregnancy, the second half of pregnancy and postpartum period.

Blood samples from animals were obtained in the morning hours before feeding from the ear vein into polypropylene tubes containing a blood clotting activator. Quantitative activity determination of aspartate aminotransferase (AST), alanine aminotransferase (SGPT), glutamyltranspeptidase (GTP), total lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) was detected on a biochemical automatic analyzer ACCENT-200 with the help of reagent kits by Cormay (Poland).

Statistical data processing was carried out using the method of single-factor analysis of variance and multiple comparison of the Newman-Keils criterion in the Primer of Biostatistics 4.03 program for Windows, on an IBM-compatible computer. The differences were considered reliable at $p \leq 0.05$. The obtained digital data were analyzed using the statistical method of one-factor analysis of variance "Biostatistics 4.03" for Windows. Reliable differences were considered when $P \leq 0.05$.

3 Results

Considering that the determination of blood enzyme systems is a sensitive and subtle indicator of pathological processes in the body, we analyzed the activity of such indicators, as aspartate aminotransferase (AST), alanine aminotransferase (SGPT), glutamyltranspeptidase (GTP), total lactate dehydrogenase (LDH) and alkaline phosphatase (ALP) (see table 1).

Table 1. Serological comparison of isoantigens in pregnant sows of different farrowing multiplicity using antiserum

№	Indicators	Non - pregnant (n=10)	1st half of pregnancy (n=10)	2nd half of pregnancy (n=10)	Postpartum period (n=10)
1.	LDH, U/l	128.20±2.58*	170.80±2.99*	213.60±7.66*	134.50±4.27*
2.	GTP, U/l	21.42±0.78	34.42±1.18*	33.10±1.72	19.77±1.39*

3.	Alkaline phosphatase (ALP), U/l	47.54±1.65	53.04±1.42	82.47±2.51*	24.88±2.03*
4.	AST, U/l	49.60±1.97	43.40±1.68*	44.90±1.54	54.90±1.12*
5.	SGPT, U/l	47.32±1.45	47.42±0.96	75.91±0.88*	47.08±0.52*
Note: the statistical significance of the differences (p≤0.05) with an earlier date is indicated *					

The data in Table 1 show that the activity of AST increased most during the postpartum period and exceeded the analyzed periods by 9.2%, 20.9%, 18.2 % (P<0.05) and was almost unchanged. SGPT in the second half of pregnancy increased by 37.9% (P<0.05), and in the postpartum period it decreased almost to the initial level as in non-pregnant animals.

GTP level the most increased in the first half of pregnancy and amounted to 34.42±1.18 (P<0.05) and in the postpartum period, it significantly decreased to 19.77±1.39 U/l. ALP activity increased intensively throughout the pregnancy period. At the same time, the highest value (P<0.05) of alkaline phosphatase was in the second half of pregnancy - 82.47±2.51 U/l.

The activity of total LDH increased only during pregnancy, exceeding the level of non-pregnant individuals by 39.9% and in the postpartum period by 37.0% (P<0.05).

The conducted biochemical studies of blood serum indicate that the state and direction of carbohydrate and protein metabolism in animals change significantly. The most significant changes are observed in the second half of pregnancy.

The results of the crude protein study are presented in Table 2. It was revealed that the level of crude protein in non-pregnant and in pregnant sows in different periods does not differ significantly (73.02±1.27 g/l; 78.58±0.64 g/l), and the differences between the groups during pregnancy are statistically significant (87.86±1.20; 81.69±1.05). The crude protein content in the blood serum of sows in gestation period is higher than the value of this indicator in non-pregnant individuals and in postpartum period on average by 10.5% and 11.8% respectively.

Table 2. Results of the study of blood sera of test groups piglets in the long-term complement binding assay

№	Indicators	Non - pregnant e (n=10)	1st half of pregnancy (n=10)	2nd half of pregnancy (n=10)	Postpartum period (n=10)
1.	Crude protein, g/l	73.02±1.27	87.86±1.20*	81.69±1.05*	78.58±0.64
2.	Albumins g/l	39.30±0.04	48.24±1.17	39.56 ±1.02	38.04 ±0.13
3.	Globulins, g/l	33.72±0.08	39.62±0.22	42.13±0.09	40.54±0.04
4.	Albumins/ Globulins ratio	1.16±0.09	1.22±0.18	0.93±0.09	0.94±0.11
5.	Urea, mmol/l	4.35±0.10	5.11±0.13*	7.31±0.21*	2.94±0.04*
6.	Creatinine, mmol/l	112.50±1.49*	136.60±2.10*	108.00±1.88*	145.30±2.56*
Note: the statistical significance of the differences (p≤0.05) with an earlier date is indicated *					

Studying the content of protein fractions, we paid attention to the protein coefficient, i.e. the ratio of albumins to globulins. The highest ratio of albumins to globulins in sows was found in the first half of pregnancy – 1.22±0.18. This is probably due to a decrease in the level of albumins and an increase in the level of globulins. In the second half of pregnancy, there is an intensive growth of the fetus and the albumin-globulin ratio indicates the mobilization of the mother's protein reserves. In postpartum period, the albumin-globulin ratio in sows decreases. This fact indicates that sows have a faster restructuring in protein metabolism as a result of pregnancy.

Throughout the entire period of research, a wave-like change in the concentration of creatinine in the blood serum was established. By the end of pregnancy, its level had decreased to 108.00±1.88 mmol/l. Obviously, this is proof that the decrease in crude protein was mainly due to albumin fractions, the number of which decreased in the second half of pregnancy to 39.56±1.02 g/l and continued to decline in the postpartum period, reaching 38.04±0.13 g/l ($P<0.05$). We associate this change with the applied vaccination scheme on the farm, which indicates an increase in the content of globulin fractions per antigen injection due to the production of specific antibodies.

Studies have also been conducted to analyze the content of crude protein, urea and creatinine in the blood serum of sows before pregnancy, during pregnancy and in postpartum period. As a result of the research, it was found that significant differences in the level of crude protein in the sow blood serum were detected between the first and second halves of pregnancy. The lowest crude protein content was found in the second half of pregnancy – 47.5±4.15 g/l. Thus, this period is critical for the fetoplacental relationship.

When studying the urea content, significant differences were found in sows in the first and second half of pregnancy – there was a tendency of a relative increase of 30.1% in relation to the first half of pregnancy. In postpartum period there was an intensive decrease to 2.94± 0.04 mmol/l, which is 59.8% lower than the second half of pregnancy.

The creatinine level in the studied animals for the entire study period had a wave-like character with a significant change in its level. So the highest value was in postpartum period 145.30±2.56 mmol/l, which is 22.5%, 5.9%, and 25.7% ($p<0.05$) higher than similar indicators in the observed periods.

The peculiarities of neonatal development of piglets belonging to different experimental groups are compared with the level of isoimmunized mothers. The results are shown in Table 3.

The presented results show that the maximum number of piglets of experimental groups was born from sheep with titers of 1:40, which was 53% of all newborns. Offspring from sheep with titers 1:20 are 32.9% of the total number of piglets obtained, and in sheep with titers 1:10, offspring respectively amounted to 14.1%.

4 Discussion

Extremely complex connections were revealed between immunocompetent cells of both identical and different specificity. Depending on the dose and characteristics of the antigen, the number of individual subpopulations of lymphoid cells, the degree of their differentiation, and specific conditions, the immune response is either enhanced, limited, or qualitatively modified.

Table 3. Results of the study of blood sera of control group piglets in the long-term complement binding assay

Piglet groups		Maternal antibody titers before childbirth		
		1:10	1:20	1:40
Group 1	newborns	-	8	49
	postnatal losses	-	6	18
Group 2	newborns	3	16	85
	postnatal losses	-	7	14
Group 3	newborns	56	114	89
	postnatal losses	2	3	2

An important addition to the studies conducted was the analysis of the distribution of piglets in groups depending on the seropositivity of mothers. So, in the first group, 86% were born from sows with titers 1:40, 14% - from sows with titers 1:20.

In 81.7% of piglets of the second group of mothers had isoantibodies titers 1:40, in 15.4% - 1:20, and piglets whose mothers had titers 1:10 were 2.9%.

In the third group, the distribution of newborns by the degree of isoimmunization of sows is presented as follows: piglets born from sows with titers 1:40 amounted to 34.4%, newborns. Newborns, whose mothers had 1:20 titers before the eye, accounted for 44%. The rest were born from sows with titers of 1:10, which corresponded to 21.6%.

Therefore, the nature of the detected changes depends on the level of antigenic load of the mother's body during pregnancy; these changes are the reason for the high risk of isoimmunization in the offspring. The established pathological signs in the placental structure reveal the mechanism of isoimmunization effect in newborn piglets and underlie the further level of their viability (see table 4).

It was found that most aborted fetuses during this period of pregnancy were long-term assigned to the first group. This is 24 fetuses - 37.4% of the total number of abortions for the entire period of prenatal development for all experimental groups. The registration of abortions in this group in the pre-birth period took place on the 34-37 day - 15 abortions, on the 41- 43 day - 9 abortions.

Part of the aborted fruits made up the second group. Their number is lower than in the first group - 8 fetuses (12.1% of the total number of abortions for the entire period of prenatal development for all experimental groups). Abortions in this group were observed for 30-46 days, 3 of which occurred on 30-35 days of embryogenesis and 5-43-46 days.

No abortions were observed in the third group. In the control group during embryogenesis (group 4), 5 fetuses were aborted (41.6% of the total number of abortions observed in isoimmunized sows) for 30-35 days and two fetuses (16.8%) - 41-43 days.

Table 4. Functional indicators of animals

Aborted fruit	Prenatal age, days	Sets:Lidina (cm), massa (g)	
Pre-pregnancy period			
Group 1	34-37	massa	1.0±0.16*
		lidina	1.9±0.24*
	41-43	massa	5.4±0.35*
		lidina	3.6±0.15*
Group 2	30-35	massa	1.1±0.12*
		lidina	2.1±0.31*
	43-46	massa	7.8±0.28*
		lidina	4.2±0.15*

Group3 (control)	30-35	massa	1.8±0.21
		lidina	2.8±0.18
	41-43	massa	9.8±0.08
		lidina	5.1±0.31
The fetal period			
Group 1	95-100	massa	540.0±0.24*
		lidina	23.5±0.54*
	117-120	massa	1100.0±0.23*
		lidina	27.9±1.17*
Group 2	90-94	massa	540.0±0.35*
		lidina	24.5±0.64*
	117-120	massa	1600.0±0.21*
		lidina	32.8±0.42*
Group3 (control)	90-95	massa	800.0±0.10
		lidina	25.7±0.30
	114	massa	2500.0±0.15
		lidina	35.4±0.23

5 Conclusions

We consider an increase in the activity of transaminases in blood serum as an early sign of hepatic cytolytic syndrome, indicating a violation of the protein-synthesizing function of the liver. An increase in the activity of GTP and ALP indicates a violation of the outflow of bile and the accumulation of its components in the liver and blood. An increase in the activity of total LDH in blood serum indicates the activation of glycolysis processes and is closely related to the functional state of liver cells. The dynamics of changes mainly indicates the state of the animal's organism as a whole, its immunological restructuring, the formation of antibodies.

Thus, based on the data obtained, we believe that the activity of blood enzyme systems has the greatest critical effect in the second half of pregnancy, which is noticeable in the phase change in the level of enzymes causing the transformation of homeostasis

The obtained data can be used as constant data for pigs in the following periods of intensive rearing: before pregnancy, the first and second half of pregnancy and postpartum period.

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