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Biochemical serum characteristics in fattening pigs infected with porcine circovirus type 2

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Abstract. Circoviral infection is one of the most widespread disease among pig populations which cause significant economic losses. Plural complications, differences in biology of known strains, wide range of clinical and biochemical changes require the detail study of its pathogenesis. The aim of the study was to identify biochemical alterations in commercial pig herds caused by circoviral infection progress. The study was carried out with use spontaneously infected animals selected from the farming pig herd aged 17-21 weeks. Animals were previously vaccinated with various vaccines and protocols. Viremia was confirmed by RT-PCR method that serves as the main criterium to divide animals for control and experimental groups. The blood samples were collected to detect biochemical parameters with applying an automated biochemical analyzer, high-performance liquid chromatography and inductively coupled plasma emission spectrometry methods. The most higher circovirus concentration observed for 17 weeks of age with consequently reciprocal decrease at 18 weeks and an increase in PCV2 DNA concentration at 19-21 weeks, while it remained significantly consistent during 19-21 weeks. The progress in circoviral infection likely led to a notable increase in total protein content in blood serum at 17th (by 11.6%; P<0.05), 19th (by 12.7%; P<0.05), 20th (by 14.7%), and 21st (by 9.1%) weeks of age. These changes in animal blood from 17 to 20 weeks were attributed to globulins. The globulins content was observed in PCV2 infected pigs at 17, 18, 19, and 20 weeks exceeded control values by 34.4%, 17.7%, 18.4%, and 11.6%, respectively (P<0.05). At 21 weeks, pigs with PCV2 viremia showed a 20.9% higher (P<0.05) level of inorganic phosphorus and nearly twofold higher (P<0.05) alkaline phosphatase activity in blood serum. The selenium concentration in 17-week-old pigs in PCV2 infected group was higher by 17.8% (P<0.05) in respect with control. Contrary, decrease in vitamin E level by 23.2% (P<0.05) and 31.5% (P<0.05) was detected in pig groups for 19 and 21 weeks respectively. The primary biochemical changes in porcine circovirus infection include an increase in total protein content and globulin fractions that accompanied by a decrease in vitamin E level. These findings reflect the impact of PCV-2 infectious progress in swine homeostasis disturbance and can be applied to advance biochemical diagnostics and to construct new therapeutic and preventive measures.

Keywords: circovirus type 2 infection; viremia; blood serum; total protein; globulins; vitamin E

Біохімічні показники сироватки крові у відгодівельних свиней за цирковірусної інфекції

Анотація. Цирковірусна інфекція є однією з найбільш поширених серед поголів'я свиней, що наносить значних економічних збитків та потребує подальшого вивчення патогенезу та, зокрема, клініко-біохімічних змін. Метою роботи було встановити біохімічні зміни у свиней товарного стада на тлі розвитку цирковірусної інфекції. Дослідження проведено на спонтанно інфікованих тваринах товарного стада у віці 17-21 тижня, попередньо вакцинованих різними способами. Наявність віремії підтверджувалася методом ПЛР-РЧ, після чого тварин поділяли на дві групи – контрольну та дослідну та відбирали у них зразки сироватки крові. Визначення біохімічних показників у ній проводили з використанням автоматичного біохімічного аналізатору, а також методів високоефективної рідинної хроматографії та емісійної спектрометрії з індуктивно-зв'язаною плазмою. Найвищою концентрація вірусу була на 17 тижні життя, з подальшим зниженням на 18 тижні і наростанням концентрації ДНК ЦВС-2 на 19-21 тижнях, де вона суттєвим чином не відрізнялась. Перебіг цирковірусної інфекції призводив до вірогідного підвищення вмісту загального білка у сироватці крові на 17-му (на 11,6 %; Р<0,05), 19-му (на 12,7 %; Р<0,05), 20-му (на 14,7 %) і 21-му (на 9,1 %) тижнях життя. Такі зміни у тварин віком від 17 до 20 тижня життя відбувались за рахунок глобулінів, рівень яких у дослідних свиней 17-ти, 18-ти, 19-ти і 20-ти тижневого віку перевищував контрольні значення на 34,4%; 17,7%; 18,4% і 11,6 % відповідно (Р<0,05). У 21-тижневому віці в сироватці крові свиней з віремією ЦВС-2 було встановлено вищий на 20,9 % (P<0,05) рівень неорганічного фосфору та майже у 2 рази (P<0,05) – активність лужної фосфатази. Концентрація селену у 17-тижневих свиней на відгодівлі у дослідній групі була вищою на 17,8 % (Р<0,05), тоді як у 19- і 21-тижневому віці було виявлено вірогідне зниження рівню вітаміну Е на 23,2 % (P<0,05) та 31,5 % (P<0,05) відповідно. Основними змінами біохімічних показників за цирковірусної інфекції свиней слід вважати наростання рівню загального білка та глобулінових його фракцій на тлі зниження вмісту вітаміну Е, що відображає вплив інфекційного процесу ЦВС-2 на організм свиней і може бути використано для біохімічної діагностики та розробки лікувально-профілактичних заходів.

Ключові слова: цирковірусна інфекція свиней 2 типу; віремія; сироватка крові; загальний білок; глобуліни; вітамін Е

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Introduction

Certain viral infections in pigs are a significant factor that leads to substantial economic losses in the swine industry. Progressive losses in indicators of productivity have recently occurred in farms associated with cases of circoviral infections (Poulsen Nautrup et al., 2023). PCV2 and its associated syndromes are one of the most widespread problems in infectious pig pathology worldwide, as well as in Ukraine, since they have been discovered (Gillespie et al., 2009; Masiuk et al., 2015).

Porcine Circovirus 2 (PCV2) is an icosahedral, single-stranded circular DNA virus without an envelope, classified within the family Circoviridae, genus Circovirus (Allan and Ellis, 2000). This genus includes PCV1, PCV2, and PCV3, among which PCV2 infects pigs and leads to various manifestations such as post-weaning multisystemic wasting syndrome (PMWS), porcine dermatitis and nephropathy syndrome (PDNS), and reproductive failure (Phan et al., 2016).

Nearly all members of the Circoviridae family exhibit lymphotropic and, consequently, immunopathogenic properties. Circovirus Type II directly infects macrophages, dendritic cells in peripheral lymphoid organs, and lymphocytes. Furthermore, the pathogen possesses properties that enable prolonged persistence in antigen-presenting cells (macrophage) (Wellenberg et al., 2000). The marked weight loss in clinically affected pigs with circoviral infection may be attributed to the impairment or blockade of lymphocyte immune function, associated with the regulation of processes related to the growth and regeneration of somatic tissues (Ladekjaer-Mikkelsen et al., 2002).

The pathogenesis of Porcine Circovirus Type II infection is complex and not fully understood. PCV2 replicates in actively dividing cells, including lymphocytes, macrophages, endothelial, and epithelial cells. PCV2 infection leads to a disease known as PCV2-associated disease (PCVAD), which encompasses systemic forms such as post-weaning multisystemic wasting syndrome (PMWS), porcine dermatitis and nephropathy syndrome (PDNS), porcine respiratory disease complex (PRDC), proliferative and necrotizing pneumonia (PNP), and enteric disease (ED) (Meng, 2012; Segalés, 2012).

One of the most effective strategies for controlling circoviral infection is the administration of PCV2 vaccines to piglets or sows before farrowing. It has been demonstrated that vaccinating sows reduces the prevalence of PCV2 viremia and enhances the productivity of their offspring (Pleguezuelos et al., 2021; Poulsen Nautrup et al., 2023). Studies indicate that vaccinating piglets against PCV2 significantly reduces the virus load in their blood. This, in turn, is associated with a reduction in clinical signs and an improvement in productivity (Figueras-Gourgues et al., 2019; Woźniak et al., 2019).

PCV2 affects the functioning of organs and systems, leading to morphological and biochemical changes in the blood composition (Rajesh et al., 2020). Meanwhile, the biochemical profile of animals in the industrial herd is utilized to monitor the health status of widely used animals, assessing pig herds for the presence of diseases, predicting their productivity, or evaluating feeding practices (Yefimov et al., 2017; Zhang et al., 2022; Dimitrakakis et al., 2022). However, the assessment of these parameters in healthy industrial pigs is seldom encountered. Data on biochemical changes during circoviral infection in commercial pig herds under PCV2 immunoprophylaxis are absent in the accessible literature. The study of this issue will contribute to enhancing our understanding of specific aspects of the disease's pathogenesis and elucidate characteristic pathognomonic changes in the blood's biochemical composition during circoviral infection in pigs.

Therefore, the purpose of this study was to identify the biochemical changes in commercial pig herds in the context of the development of circoviral infection.

Materials and methods

Animals. The research was carried out on the industrial pig farming complex in Ukraine, considering the fact that circovirus infection was more frequently identified in the studied farm compared to other swine producers. Monitoring was carried out on swine aged 17-21 weeks to assess mortality associated with PCV2 infection during the autumn period. An increase in the number of anemic animals and a rise in the mortality rate were observed at this age. PCR investigations confirmed the presence of PCV2. Animals selected from this facility were included in the presented study as the PCV2-positive group. An analysis of autopsy results for animals that died during monitoring revealed the presence of stomach ulcers in 22% of deceased animals in the experimental group, while the control group had a rate of 11%. The mortality rate during this period was 1.5% higher in animals infected with PCV2.

The animals were housed in group pens according to zoohygienic requirements. Both the control and experimental groups of animals received nutritionally and granulometrically identical for the complete feed.

Piglets in the control group were intramuscularly vaccinated on day 30 of life with a commercial inactivated vaccine against Porcine Circovirus Type 2 at a dose of 0.5 ml. Piglets in the experimental PCV-2 positive group were administered a commercial vaccine against enzootic pneumonia and PCV2 intradermally on day 30 of life, with the vaccine containing Mycoplasma hyopneumoniae expressing the capsid protein of porcine circovirus type 2a, inactivated, at a dose of 0.2 ml.

For the study, blood samples were collected from five experimental and control pigs in each age group, taking into the account the requirements of Ukrainian Law No. 3447-IV dated February 21, 2006, «On the Protection of Animals from Cruelty,» and aligned with the fundamental principles of the «European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes» (Strasbourg, 1986).

Laboratory research was conducted at the Research Center for Biosecurity and Environmental Monitoring of Agricultural Resources AIC at the Dnipro State Agrarian and Economic University in Dnipro.

Blood sample collection. Blood samples of 5 ml were collected from four animals in each age group through venipuncture of the anterior vena cava, using test-tubes with a clot activator. After centrifugation at 1800 rpm for 10 minutes, serum was separated and stored at -20 °C until analysis.

PCR detection of PCV2. For PCR analysis of PCV-2, samples were pooled to provide an objective assessment of the viral load in the herd (Nielsen et al., 2018). A total of 100 μ L of serum was collected from every individual sample and combined to create a 200 μ L pooled sample which was used for PCR analysis. Genomic DNA from the serum samples was extracted with using the commercial BIOEXTRACT «SUPERBALL» kit (Biosellal, France) and the automated nucleic acid extraction station KingFisher Duo (USA).

The «EXOone PCV2» test kit from EXOPOL (Spain) was applied for real-time detection of Porcine Circovirus Type 2 DNA. Amplification and result detection were conducted on the BioRad CFX 96 instrument (USA). The obtained results were shown in genome equivalents (g.e.) of the virus per 1 ml of material. A positive control sample (calibrator) with a known concentration included in the diagnostic kit was used as a standard.

Serum biochemical parameters measuring. The biochemical serum parameters were measured using commercial reagent kits. Spinreact kits (Spain) were used for measuring total protein, albumin, creatinine, urea, aspartate aminotransferase activity, alanine aminotransferase activity, alkaline phosphatase; Cormay (Poland) – for calcium, glucose; Dialab (Austria) – for inorganic phosphorus, with the assistance of the automated biochemical analyzer Miura-200 (Italy).

Age, weeks	Group	Virus content
17	control	Not detected
	PCV2 positive	1.66*107
18	control	Not detected
	PCV2 positive	2.82*105
19	control	Not detected
	PCV2 positive	$1.64^{*}10^{6}$
20	control	Not detected
	PCV2 positive	7.98*105
21	control	Not detected
	PCV2 positive	$3.46*10^{6}$

Table 1 – The content of PCV2 genetic material in serum of pigs,g.e.

The measuring of the vitamin composition of blood serum was conducted on the liquid chromatograph Agilent Technologies 1260 Infinity II (USA), using Sigma-Aldrich (France) eluents following the methodology previously described (Yefimov & Sofonova, 2014).

The mineralization of 500 µL serum samples was performed in the microwave oven Multiwave GO Plus (Anton Paar, Austria) using the EPA Method 3052. The obtained samples were analyzed for Se content using an inductively coupled plasma optical emission spectrometer Agilent 5110 ICP-OES (USA). Measurements were conducted at the characteristic emission wavelength for Se -196.026 nm. The measurements were performed on the Agilent 5110 Synchronous Vertical Dual View (SVDV) ICP-OES system, equipped with the Multi-Mode Sample Introduction System (MSIS) and the SPS 4 autosampler. MSIS operated in a dual mode with a sample introduction system consisting of a SeaSpray nebulizer with an internal diameter of 1.8 mm and an injector torch. All solutions were prepared using ultrapure water, and 65% nitric acid (Sigma-Aldrich, USA) was used for oxidation. The calibration curve was constructed using Agilent Technologies standards at 10000 µg/ml in 5% HNO₃.

The results were statistically processed using the specialized software Statistica 6 (StatSoft Inc, USA). Significance of the differences between control and PCV-positive groups was assessed by the Student's t-test. The design of data presentation was arranged with next designations: M - mean; m - standard error of the mean (SEM). The data presentation design included the following notations: M - mean; m - standard error of the mean (SEM). Changes were considered statistically significant as P < 0.05.

Table 2 – Protein metabolism characteristics in serum of pig groups for control and infected with PCV2 ($M \pm m, n = 4$)

	100 1	
Parameters	Groups	
	control	PCV2 positive
	17 weeks age	
Total protein, g/l	60.50 ± 1.29	$67.50 \pm 1.55*$
Albumin, g/l	36.80 ± 1.56	35.75 ± 4.11
Globulin, g/l	24.00 ± 1.73	$32.25 \pm 5.11*$
A/G ratio	1.58 ± 0.16	1.29 ± 0.27
	18 weeks age	
Total protein, g/l	66.75 ± 3.42	74.25 ± 2.59
Albumin, g/l	31.75 ± 1.25	32.50 ± 1.04
Globulin, g/l	35.25 ± 2.56	$41.50 \pm 1.90*$
A/G ratio	0.91 ± 0.05	0.80 ± 0.07
	19 weeks age	
Total protein, g/l	73.00 ± 2.92	82.25 ± 2.14*
Albumin, g/l	32.00 ± 0.82	34.00 ± 0.71
Globulin, g/l	40.75 ± 2.46	$48.25 \pm 2.81*$
A/G ratio	0.83 ± 0.04	0.71 ± 0.06
	20 weeks age	
Total protein, g/l	66.25 ± 1.80	$76.00 \pm 3.11*$
Albumin, g/l	37.50 ± 0.65	38.00 ± 0.71
Globulin, g/l	28.00 ± 2.48	$38.25 \pm 2.39*$
A/G ratio	1.33 ± 0.15	1.00 ± 0.04 *
	21 weeks age	
Total protein, g/l	68.50 ± 1.85	$74.75 \pm 1.89*$
Albumin, g/l	32.25 ± 1.49	$37.75 \pm 1.11*$
Globulin, g/l	36.75 ± 2.59	36.50 ± 2.22
A/G ratio	0.89 ± 0.09	1.03 ± 0.07

Results

The results of the study using the PCR method showed that the serum of pigs aged 17-21 weeks contains the PCV2 virus at various concentrations (Table 1).

The highest concentration of the virus was detected at 17 weeks of age, followed by a wave-like pattern. There was a decrease at 18 weeks and an increase in the DNA concentration of PCV2 at 19-21 weeks, where it did not significantly differ. Thus, viremia of PCV2 was established in animals of the experimental group of different ages.

The course of circovirus infection in the body of fattening pigs had a certain impact on the content of total protein in the blood serum, as well as its albumin and globulin fractions (Table 2).

Higher levels of total protein were observed among the investigated indicators of protein metabolism. Specifically, these changes were significant at the 17th (by 11.6%; P < 0.05), 19th (by 12.7%; P < 0.05), 20th (by 14.7%), and 21st (by 9.1%) weeks of life. At the same time, a similar trend was observed in animals at the age of 18 weeks.

A more detailed analysis of the levels of the main protein fractions (albumins and globulins) provides grounds to assert that the increase in its content in animals aged 17 to 20 weeks occurred due to globulins. In particular, in PCV2-positive pigs at 17, 18, 19, and 20 weeks of age, it exceeded control values by 34.4%, 17.7%, 18.4%, and 11.6%, respectively (with significance at P < 0.05). However, with increasing age, the difference between the groups gradually decreased, and in 21-week-old animals, no significant difference in the content of globulin fractions was found. On the othe groupsr hand, a higher content of albumins was detected (by 17.0%, with P < 0.05) in the oldest pigs with circovirus infection compared to animals without viremia.

At the same time, we did not observe significant differences in the metabolite levels of nitrogen metabolism including the content of urea and creatinine in the blood serum (Table 3).

In most age groups (17-, 19-, 20-, and 21-week-old), there was observed a tendency for an increase in urea content in PCV2positive animals. However, due to the significant individual variability of this biochemical parameter, no statistically differences were observed. No significant differences were found among the indicators of calcium-phosphorus metabolism in animals from both the control and experimental groups aged 17-20 weeks (Table 4).

However, at the age of 21 weeks, fattening pigs with PCV2 viremia exhibited a 20.9% higher level of inorganic phosphorus (P < 0.05), and the alkaline phosphatase activity was almost twice as much (P < 0.05).

The course of the infectious process in the body during various diseases leads to intensified processes of peroxidation. Some variations were noted in the levels of vitamins A and E, as well as selenium during the assessment (Table 5).

In particular, the selenium concentration in 17-week-old fattening pigs in the PCV2 positive group was higher by 17.8% (P < 0.05) compared to the control animals. Meanwhile, the levels of vitamins A and E did not show significant differences at this age. No differences in these indicators were observed at 18 weeks. However, at 19 and 21 weeks, there was a decrease in the level of vitamin E by 23.2% (P < 0.05) and 31.5% (P < 0.05), respectively. Thus, some changes in the non-enzymatic antioxidant defense during the course of circovirus infection in pigs were identified.

Discussion

PCV2-associated diseases lead to changes in the functioning of various organs in pigs. Considering this, alterations in the blood biochemical composition during circovirus infection can be used for indirect PCV2 diagnostics. Such studies also allow to discover specific aspects of the pathogenesis of this disease. This information is particularly valuable for herds that have undergone vaccination, as the disease may have a subclinical course as a result.

Intradermal needle-free vaccination is currently considered an alternative immunization method. It significantly reduces the level of stress and pain reactions in piglets, improving their well-being (Temple et al., 2020). Suh and Chae (2022) show that intradermal vaccination of piglets provided effective protection against experimental infections of M. hyopneumoniae and PCV2d. The results obtained in our study have shown that PCV2-positive animals exhibit PCV2 viremia after vaccination. Contrary, there was no detected PCV2 viremia in the experimental group. The

Table 3 – The content of nitrogen metabolites in serum of pig groups for control and infected with PCV2 ($M \pm m, n = 4$)

Parameters	Groups	
	control	PCV2 positive
	17 weeks age	
Urea, mmol/l	4.56 ± 0.75	4.93 ± 0.35
Creatinine, µmol/l	66.00 ± 1.87	70.00 ± 7.25
	18 weeks age	
Urea, mmol/l	4.63 ± 0.24	3.85 ± 0.46
Creatinine, µmol/l	72.75 ± 1.44	77.50 ± 1.94
	19 weeks age	
Urea, mmol/l	4.35 ± 0.46	4.80 ± 0.24
Creatinine, µmol/l	71.38 ± 4.12	68.50 ± 7.56
	20 weeks age	
Urea, mmol/l	3.66 ± 0.06	3.95 ± 0.27
Creatinine, µmol/l	70.00 ± 2.16	69.75 ± 2.72
	21 weeks age	
Urea, mmol/l	3.60 ± 0.28	3.83 ± 0.19
Creatinine, µmol/l	70.00 ± 1.35	74.00 ± 1.08

Parameters	Group	
	control	PCV2 positive
	17 weeks age	
Calcium total, mmol/l	2.84 ± 0.06	2.68 ± 0.14
Phosphorus inorganic, mmol/l	4.44 ± 0.29	4.60 ± 0.51
Alkaline phosphatase, U/l	78.26 ± 7.00	96.00 ± 18.30
	18 weeks age	
Calcium total, mmol/l	2.75 ± 0.10	2.95 ± 0.06
Phosphorus inorganic, mmol/l	3.83 ± 0.11	3.65 ± 0.18
Alkaline phosphatase, U/l	78.25 ± 17.92	66.63 ± 11.84
	19 weeks age	
Calcium total, mmol/l	2.75 ± 0.10	2.80 ± 0.11
Phosphorus inorganic, mmol/l	$3,\!00\pm0,\!04$	3.13 ± 0.09
Alkaline phosphatase, U/l	44.55 ± 4.40	50.98 ± 6.15
	20 weeks age	
Calcium total, mmol/l	2.55 ± 0.07	2.70 ± 0.06
Phosphorus inorganic, mmol/l	3.53 ± 0.15	3.68 ± 0.06
Alkaline phosphatase, U/l	64.63 ± 4.09	68.45 ± 1.64
	21 weeks age	
Calcium total, mmol/l	2.50 ± 0.04	2.60 ± 0.05
Phosphorus inorganic, mmol/l	3.50 ± 0.26	$4.23 \pm 0.27*$
Alkaline phosphatase, U/l	42.60 ± 1.68	$84.75 \pm 16.23*$

Table 4 – Parameters of calcium and phosphorus metabolism in serum of pig groups for control and infected with PCV2 ($M \pm m, n = 4$)

Table 5 –Antioxidant contents in serum of pig groups for control and infected with PCV2 ($M \pm m, n = 4$)

Parameters	Group	
Parameters	control	PCV2 positive
	17 weeks age	
Vitamin A, µg/dl	29.08 ± 2.94	27.39 ± 1.47
Vitamin E, mg/l	1.86 ± 0.30	1.47 ± 0.17
Selenium, µg/l	91.71 ± 4.71	108.02 ± 5.24 *
	18 weeks age	
Vitamin A, µg/dl	32.34 ± 3.21	31.32 ± 1.53
Vitamin E, mg/l	1.66 ± 0.13	1.63 ± 0.22
Selenium, µg/l	99.07 ± 4.74	97.02 ± 3.80
	19 weeks age	
Vitamin A, µg/dl	25.89 ± 2.03	$25,87 \pm 2,26$
Vitamin E, mg/l	1.38 ± 0.10	$1,06 \pm 0,10*$
Selenium, µg/l	103.68 ± 3.90	99.51 ± 5.01
	20 weeks age	
Vitamin A, µg/dl	30.12 ± 2.28	29.69 ± 2.55
Vitamin E, mg/l	1.76 ± 0.26	1.57 ± 0.13
Selenium, µg/l	94.73 ± 3.37	98.06 ± 4.55
	21 weeks age	
Vitamin A, µg/dl	28.88 ± 3.25	32.27 ± 2.93
Vitamin E, mg/l	2.41 ± 0.26	$1.65 \pm 0.17*$
Selenium, µg/l	97.75 ± 4.55	102.62 ± 4.91

production of antibodies against PCV2 infection does not guarantee complete protection, therefore, viremia may still occur in animals (Sibila et al., 2004). Additionally, pigs with PCV2-associated diseases often have low levels of neutralizing antibodies (Meerts et al., 2006). The presence of viremia in the experimental group may be explained by lower levels of neutralizing antibodies against PCV2 due to the use of intradermal vaccination.

The level of total protein in blood serum is determined by the combined concentration of its various fractions (Tóthová et al., 2021). Albumin is one of the most common plasma proteins, and its level is typically used as an indicator of liver or kidney function (Carvalho and Machado, 2018; Friedman and Fadem, 2010). The higher levels of albumins were observed in 21-week-old pigs infected with PCV2. This result may be explained by reduced utilization of albumins in the biosynthesis of peripheral tissues due to decreased weight gains. There was considered that the enhanced synthesis of albumins in the liver also serves as a means to preserve essential amino acids from degradation (De Feo et al., 1992).

Among different globulin fractions, the levels of α - and β -globulins remain stable after the neonatal period, with γ -globulins being the most labile fraction (Miller et al., 1961). PCV2 viremia is accompanied by an increase in the concentration of globulin protein fractions in almost all age groups. Similar results regarding porcine circovirus infection were also reported by Phaneuf et al. (2007) and Rajesh et al. (2020) in their studies. This might be a consequence of enhanced synthesis of γ -globulin protein fractions in the swine's body, particularly due to the action of secondary pathogens of bacterial origin.

It is known that PCV2 disrupts immune regulation and induces immunosuppression in pigs (Shi et al., 2021), leading to a decrease in lymphocyte count and an increase in the percentage of neutrophils in the blood (Golinar Oven et al., 2022). Because of changes in immune function, the secretion of IL-6 and IL-8 is enhanced. Their pro-inflammatory effect is due to co-infection with bacterial pathogens (Wang et al., 2020). These interleukins may play a significant role in the development of gastric ulcers associated with H. pylori, as observed in the group of pigs with PCV2 viremia.

Many authors reported an increase in non-protein nitrogen compounds in the blood serum of pigs infected with PCV2, including urea and creatinine (Wellenberg et al., 2004; Phaneuf et al., 2007). At the same time, the animals they described developed dermatitis and nephropathy syndrome. In our study, clinical signs such as dermatitis and nephropathy were not observed in pigs with viremia, while azotemia was common for pigs with kidney lesions of various origins (Marin et al., 2018; Zheng et al., 2020).

There was a decrease in the level of tocopherol in the blood serum in pigs of different ages with viremia. Vitamin E is known for its antioxidant properties, implemented through nonenzymatic ways, including a direct reaction with active oxygen forms (Miyazawa et al., 2019). On the other hand, viral infections, including PCV2, are characterized by the development of oxidative stress (Checconi et al., 2020; Rajesh et al., 2020). The changes identified in our study may indicate the consumption of vitamin E to neutralize the formed active oxygen species. Meanwhile, an increase in the selenium level was observed in one of the age group. Selenium is known to be part of the components of the enzymatic antioxidant defense system (Roy et al., 2005). Perhaps, these changes are the consequence of a compensatory reaction to the virus replication in the context of sufficient selenium supply in pigs.

Conclusion

Despite vaccination, PCV2 viremia was observed at 17-21 weeks in the pigs of the experimental group. No typical clinical signs were observed in these animals. However, certain changes in

biochemical parameters were identified. The main change was an increase in the level of total protein due to its globulin fractions, reflecting the immune response of animals to the secondary bacterial pathogens. However, azotemia was not observed as the result of subclinical course of the infectious process. At the same time, a decrease in the content of vitamin E was established as a compensatory reaction. The obtained results can be used for biochemical diagnostics and the development of therapeutic and preventive measures for PCV2 infection in pigs.

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