Chapter 6

IMMUNE SYSTEM DEVELOPMENT IN NEONATAL PIGLETS

Masiuk Dmytro, Nedzvetsky Victor, Kokariev Andrii
DOI https://doi.org/10.30525/978-9934-26-454-2-6

INTRODUCTION

The most important stage in the body development is an early postnatal adaptation[1]. This period has the highest percentage of newborn piglets deaths on the pig farms[23]. Among the main reasons are non-invasive pathology of the digestive and respiratory tract, caused by the low level of innate immunity as a result of increased technological influence, stresses of various etiologies, and violations of the sows growing technology[4, 5, 6]. The combination of all these factors leads to violations in the fetal period of the body's development, a decrease in the colostrum quality and is accompanied by newborns immunodeficiency[7].

So, the increase of the newborns immune status with the use of natural immunomodulators, which influence directly on the immune competent cells or indirectly due to changes in various biological reactions of the organism[89].

---

[1]Qi M., Tan B., Wang J., Liao S., Li J., Cui Z., Shao Y., Ji P., Yin Y. Postnatal growth retardation is associated with deteriorated intestinal mucosal barrier function using a porcine model. *Journal of cellular physiology*. 2021. Vol. 236, No. 4. P. 2631–2648.

[2]Huang G., Li X., Lu D., Liu S., Suo X., Li Q., Li N. Lysozyme improves gut performance and protects against enterotoxigenic Escherichia coli infection in neonatal piglets. *Veterinary research*. 2018. Vol. 49, No. 1. P. 20.

[3]Farmer C., Edwards S. A. Review: Improving the performance of neonatal piglets. *Animal: an international journal of animal bioscience*. 2022. Vol. 16, Suppl 2. P. 100350.

[4] Aegerter H., Lambrecht B. N., Jakubzick C. V. Biology of lung macrophages in health and disease. *Immunity*. 2022. Vol. 55, No. 9. P. 1564–1580.

[5]Holda K. O., Masiuk D. M., Kokariev A. V., Vasilenko T. O. Colostral immunity of piglets to the virus of Aujeszky's disease with active sows' immunization. *Theoretical and Applied Veterinary Medicine*. 2020. Vol. 8, No. 4. P. 257–260.

[6]Schlosser-Brandenburg J., Ebner F., Klopfleisch R., Kühl A.A., Zentek J., Pieper R., Hartmann S. Influence of Nutrition and Maternal Bonding on Postnatal Lung Development in the Newborn Pig. *Frontiers in immunology*. 2021. Vol. 12. P. 734153.

[7]Abrao Trad A. T., Buddington R., Enninga E., Duncan J., Schenone C. V., Mari G., Buddington K., Schenone M. *Report of an Experiment With a Fetal Ex-Utero Support System in Piglets. Cureus*. 2023. Vol. 15, No. 4. P. e38223.

[8]Hedges J. F., Snyder D. T., Robison A., Thompson M. A., Aspelin K., Plewa J., Baldridge J., Jutila M. A. A TLR4 agonist liposome formulation effectively stimulates innate immunity and enhances protection from bacterial infection. *Innate immunity*. 2023. Vol. 29, No. 3–4. P. 45–57.

Some immunomodulators influence the homeostatic balance of the body's systems, in particular, the immune and neuroendocrine[10]. The use of immunomodulators without side effects and which can balance the immune system with its adaptive capabilities is very important research direction[11, 12].

The most effective way of newborns immune correction is an influence through the mother's body[13, 14]. The study of congenital immunity of piglets in the early stages of postnatal adaptation with the use of immunotropic drugs in the mother-colostrum-newborn system is of particular relevance[15, 16]. Therefore, the purpose of the study was to determine the ways of increasing piglets congenital immunity in the early postnatal period of ontogenesis with the application of «EHCW».

1. Haematological and biochemical characteristics blood

Blood parameters serve as markers of physiological processes occurring in the various animals[17]. Consequently, the influence of various preparations on the swine organism is reflected in the values of blood parameters[18]. The most common blood parameters include the number of erythrocytes and

---

[9]De Carvalho R. H., Callegari M. A., Dias C. P., Kirwan S., da Costa M. C. R., da Silva C. A. Euglena gracilis β-Glucans (1,3): Enriching Colostrum of Sow for Enhanced Piglet Immunity. *Animals: an open access journal from MDPI*. 2023. Vol. 13, No. 22. P. 3490.

[10]Grün V., Schmucker S., Schalk C., Flauger B., Weiler U., Stefanski V. Influence of Different Housing Systems on Distribution, Function and Mitogen-Response of Leukocytes in Pregnant Sows. *Animals: an open access journal from MDPI*. 2013. Vol. 3, No. 4. P. 1123–1141.

[11]Chepngeno J., Amimo J. O., Michael H., Jung K., Raev S., Lee M. V., Damtie D., Mainga A. O., Vlasova A. N., Saif L. J. Rotavirus A Inoculation and Oral Vitamin A Supplementation of Vitamin A Deficient Pregnant Sows Enhances Maternal Adaptive Immunity and Passive Protection of Piglets against Virulent Rotavirus A. *Viruses*. 2022. Vol. 14, No. 11. P. 2354.

[12]Branch D. W. Physiologic adaptations of pregnancy. *Am J Reprod Immunol*. 1992. Vol. 28, No. 3–4. P. 120–122.

[13]Inoue R., Tsukahara T. Composition and physiological functions of the porcine colostrum. *Animal science journal = Nihon chikusan Gakkaiho*. 2021. Vol. 92, No. 1. P. e13618.

[14]Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677.

[15] De Carvalho R. H., Callegari M. A., Dias C. P., Kirwan S., da Costa M. C. R., da Silva C. A. Euglena gracilis β-Glucans (1,3): Enriching Colostrum of Sow for Enhanced Piglet Immunity. *Animals: an open access journal from MDPI*. 2023. Vol. 13, No. 22. P. 3490.

[16]Maciag S. S., Bellaver F. V., Bombassaro G., Haach V., Morés M. A. Z., Baron L. F., Coldebella A., Bastos A. P. On the influence of the source of porcine colostrum in the development of early immune ontogeny in piglets. *Scientific reports*. 2022. Vol. 12, No. 1. P. 15630.

[17]Lipsit S., Facciuolo A., Scruten E., Griebel P., Napper S. Plasma Cytokines and Birth Weight as Biomarkers of Vaccine-Induced Humoral Responses in Piglets. *Frontiers in veterinary science*. 2022. Vol. 9. P. 922992.

[18]Holda A. A., Yefimov V. H. Biochemical serum characteristics in fattening pigs infected with porcine circovirus type 2. *Theoretical and Applied Veterinary Medicine*. 2023. Vol. 11, No. 1. P. 15–21.

leukocytes, the level of hemoglobin, and the ratio of different leukocyte forms[19].

The results obtained in our study showed that the morpho-biochemical blood parameters were within physiological limits in piglets of the first day of life (Table 1).

Table 1

Morpho-biochemical blood parameters of piglets during early postnatal ontogenesis and their correction with the «EHCW» preparation (M±m, n = 10)

Age		Hemoglobin, g/L	Hematocrit, %	Erythrocytes, T/L	Leukocytes, G/L
0 hours	control	103,60±4,24	35,56±2,17	4,16±0,12	3,32±0,28
	«EHCW»	119,80±5,41*	41,26±1,16*	4,68±0,13*	5,00±0,55*
4 hours	control	101,00±5,23	32,70±1,15	4,13±0,08	6,70±0,72^{00}
	«EHCW»	117,00±4,18*	37,08±1,86	4,45±0,07*	11,30±1,41*
3 days	control	95,00±3,79	27,36±1,92^{0}	3,79±0,11	6,78±0,73^{00}
	«EHCW»	111,20±5,77*	30,95±0,51	4,17±0,10*	7,64±1,03
7 days	control	85,40±5,08^{0}	28,16±3,27^{0}	3,57±0,10^{0}	9,72±1,16^{000}
	«EHCW»	100,60±4,24*	31,26±1,70	3,90±0,10*	14,70±1,24*
14 days	control	92,60±6,29	32,32±0,60	3,90±0,08	8,84±0,94^{000}
	«EHCW»	102,40±5,25	35,02±1,69	4,10±0,12	8,30±0,58
23 days	control	115,20±5,80	36,44±3,43	4,62±0,17	14,76±0,99^{000}
	«EHCW»	122,00±6,77	39,18±2,57	4,82±0,15	15,38±0,69

Note: 1. 0-p≤0.05; 00-p≤0.01; 000-p≤0.001 compared to piglets before colostrum intake; 2. *-p≤0.05 compared to the control.

Three days birth later, there was observed a tendency for a decrease in the number of erythrocytes and the level of hemoglobin in the blood of piglets by 8.89% and 8.30%, respectively, compared to colostrum-deprived piglets. In addition, piglets exhibited a decrease in hematocrit level by 23.06% (p≤0.05) compared to newborn piglets.

Observed changes can be mediated by the influence of a complex of factors on the organism of newborns, including high uterine pressure during

[19] Yefimov V., Kostiushkevych K., Rakytianskyi V. Influence of feed additive from peat on morphological and biochemical blood profile of piglets. *Veterinarija ir Zootechnika*. 2017. № 75. С. 17–21.

the expulsion phase, thermal, oxygen, biological, and other factors. All of these factors act on the newborn's organism from the first moments of life, affecting all body systems, which is crucial for organism development[20, 21].

It has been determined that the lowest values of erythrocyte number and hemoglobin level were observed in piglets on the 7th day of life. During this time, these parameters likely decreased by 14.18% and 17.57% ($p \leq 0.05$), respectively, compared to the values of these parameters in newborn piglets. Compared with 3 days age animals, these parameters showed insignificant differences, at 5.81% and 10.11%, respectively. It should be noted that hematocrit in piglets at 7 days of age remained almost unchanged since the 3rd day of life and was more than 20% lower than in colostrum-deprived piglets.

Therefore, during the first week of life, piglets undergo changes in blood content based on morphological and biochemical parameters, accompanying the development of an anemic state[22]. Anemia in neonatal piglets may occur due to iron deficiency, in the context of high growth intensity of their organisms[23]. In the blood of 7–8-day-old piglets, there is a decrease in hemoglobin level, erythrocyte count, and hematocrit, mainly due to the depletion of iron stores in the piglets' bodies, in the context of intensive metabolic utilization.

Subsequently, an increase in morpho-biochemical blood parameters was observed in piglets. By the end of the second week of life, the measuring of animal blood data showed an increase in hematocrit level by 14.77%, erythrocyte number by 9.24%, and hemoglobin content by 8.41% compared to animals at one week of age. However, these parameters still remained lower by 9.11%, 6.25%, and 10.62%, respectively, compared to piglets before colostrum intake.

The piglets exhibited the highest values of erythrocyte count, hematocrit level, and hemoglobin level on the 23rd day of life, that were the highest throughout the entire study period. These values were higher than

---

[20]Dong L., Zhong X., Zhang L., Kong L., Kong Y., Kou T., Wang T. Impaired intestinal mucosal immunity is associated with the imbalance of T lymphocyte sub-populations in intrauterine growth-restricted neonatal piglets. *Immunobiology*. 2015. Vol. 220, No. 6. P. 775–781.

[21]Bidarimath M., Tayade C. Pregnancy and spontaneous fetal loss: A pig perspective. *Molecular reproduction and development*. 2017. Vol. 84, No. 9. P. 856–869.

[22]Merlot E., Pastorelli H., Prunier A., Père M. C., Louveau I., Lefaucheur L., Perruchot M. H., Meunier-Salaün M. C., Gardan-Salmon D., Gondret F., Quesnel H. Sow environment during gestation: part I. Influence on maternal physiology and lacteal secretions in relation with neonatal survival. *Animal*. 2019. Vol. 13, Suppl 7. P. 1432–1439.

[23]Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677.

in newborn piglets by 11.06%, 2.48%, and 11.20%, respectively, and exceeded the values of seven-day-old piglets by 29.41%, 29.40%, and 25.87%, respectively.

Therefore, the early postnatal period of development in piglets was characterized by relatively high values of absolute erythrocyte count, hematocrit level, and hemoglobin level after birth, significant decrease during the first week of life, and gradual increase over the next 16 days.

The changes in the number of leukocytes in piglets' blood during the perinatal period were characterized by a gradual increase in its absolute number. Within the first 4 hours after birth, the number of leukocytes in piglets' blood quickly increased by 2.02 times and remained at this level during the first 3 days of life. There was next wave of increase in the number of leukocytes on the 7th day after birth, although not as pronounced as the previous one. Obtained data have shown that the absolute leukocyte count exceeded that of 3-day-old piglets by 44.2% and nearly tripled compared to pre-colostrum intake piglets ($p \leq 0.001$) of this age group. By the end of the second week of life, the number of leukocytes in piglets slightly decreased by 9.1% compared to 7-day-old ones, but it remained significantly higher by 2.7 times ($p \leq 0.001$) than in newborn animals.

During the period from the 14th to the 23rd day of life, there was a further increase in the number of leukocytes in piglets, amounting to 67.0%. As a result, the total leukocyte count in 23-day-old piglets exceeded that of pre-colostrum intake pigs and 3-day-old piglets by more than 4 and 2 times, respectively ($p \leq 0.001$).

The neonatal period in piglets was characterized by a sharp increase in leukocyte count in the first day of life, followed by gradual growth in the peripheral blood over the next 22 days with a slight decrease at the age of 2 weeks. These changes in the mentioned morphological and biochemical parameters in piglets affected the erythrocyte indices (Table 2).

There was a tendency towards a decrease in mean corpuscular volume (MCV) by 15.03% alongside an increase in mean hemoglobin concentration (MCHC) by 20.56% in 3-day-old piglets compared to the values of the piglets at 4 hours of life. Subsequently, until the end of the second week, a reverse trend was observed, reflected in a 14.10% increase in MCV and an 18.81% decrease in MCHC compared to 3-day-old piglets. By the middle of the fourth week of life, these indices once again changed their values in the direction of the initial tendency.

It has been shown that changes in erythrocyte size during the early postnatal period negatively correlated ($r = -0.91$) with the mean hemoglobin concentration in a single erythrocyte, contributing to a relatively constant value of the mean hemoglobin content in the erythrocyte.

Table 2

Erythrocyte indices in piglets during the early postnatal ontogenesis period and their correction with the " «EHCW» " preparation (M±m, n=10)

Age		MCV, fl	MCH, pg	MCHC, %	Fi, Un.
0 hours	control	85,30±4,25	24,95±0,56	29,28±0,91	0,75±0,02
	«EHCW»	88,16±0,14	25,58±0,78	29,02±0,87	0,77±0,02
4 hours	control	79,13±4,75	24,47±1,20	30,99±1,76	0,73±0,04
	«EHCW»	83,15±3,25	26,25±0,66	31,64±0,72	0,79±0,02
3 days	control	72,48±6,12	25,11±1,25	35,30±2,89	0,75±0,04
	«EHCW»	74,23±0,83	26,60±0,86	35,86±1,29	0,80±0,03
7 days	control	78,27±7,06	23,88±1,00	31,11±2,25	0,72±0,03
	«EHCW»	80,15±3,94	25,77±0,64	32,35±1,50	0,77±0,02
14 days	control	82,92±1,28	23,71±1,34	28,66±1,95	0,71±0,04
	«EHCW»	85,31±3,11	24,93±0,89	29,25±0,73	0,75±0,03
23 days	control	79,29±8,44	24,88±0,61	32,88±4,52	0,75±0,02
	«EHCW»	81,42±5,24	25,28±0,77	31,32±1,55	0,76±0,02

Thus, the neonatal period in piglets was accompanied by wave-like changes in hematological indices. These fluctuations were manifested as a decrease in erythrocyte volume and an increase in hemoglobin concentration in the first three days of life and opposite changes in these indicators from the third day to the end of the second week of life.

The study of different types of leukocytes indicates that before colostrum intake and in the first hours of life, the neutrophil fraction dominated in piglets' blood, while the counts of eosinophils and monocytes were low (Table 3).

During the neonatal period, there was a redistribution of leukocyte fractions characterized by a gradual increase in the percentage of eosinophils and monocytes, accompanied by oscillatory changes in the neutrophil and lymphocyte fractions.

Probable changes in the relative content of leukocyte fractions were observed as early as 4 hours after birth. During this time, the percentage of neutrophil granulocytes increased by 21.0% ($p \leq 0.01$), monocytes by 85.7% ($p \leq 0.01$), and the lymphocyte fraction decreased by 33.0% ($p \leq 0.01$) compared to piglets before colostrum intake. The eosinophil content in piglets' blood remained almost unchanged during the first 4 hours of life.

Table 3

Ratio of different leukocyte forms in the blood of the control group of piglets during the early postnatal ontogenesis period (M±m, %, n=10)

Age	B	E	Neutrophils		L	M
			R	S		
0 hours	0	0,60±0,21	1,80±0,29	56,30±2,32	40,60±2,53	0,70±0,34
4 hours	0	0,70±0,34	2,70±0,60	68,10±2,39**	27,20±2,15**	1,30±0,70
3 days	0	1,20±0,29	1,20±0,38	45,80±3,71*	50,20±3,49*	1,60±0,33
7 days	0	1,70±0,29*	2,20±0,29	48,70±1,97*	45,60±1,76	1,80±0,58
14 days	0	1,10±0,48	1,60±0,37	33,80±3,80***	60,90±4,26**	2,60±0,65*
23 days	0	4,20±0,80**	1,80±0,52	49,20±2,58*	41,60±1,89	3,20±1,38

Note: * – p≤0.05; ** – p≤0.01; *** – p≤0.001 compared to piglets before colostrum intake.

The redistribution between the neutrophil and lymphocyte fractions was observed in the blood of neonatal piglets on the 3rd day of life, along with an increase in the population of eosinophils and monocytes. Compared to piglets before colostrum intake, the percentage of neutrophil granulocytes was likely lower by 18.7% (p≤0.05), while lymphocytes, on the contrary, were higher by 23.7% (p≤0.05). The level of monocytes and eosinophils increased by 2.3 and 2.0 times, respectively.

During the next 4 days of life, the ratio between neutrophils and lymphocytes remained almost unchanged, while the fractions of monocytes and eosinophils increased by 12.5% and 41.7%, respectively, compared to piglets at 3 days of life.

Analysis of the leukogram of 14-day-old piglets showed that during the same period, there was an increase in mononuclear leukocytes (lymphocytes) by 1.5 times (p≤0.01) and monocytes by 3.7 times (p≤0.05), along with a decrease in the fraction of segmented neutrophils by 40.0% (p≤0.001) compared to piglets before colostrum intake. The number of eosinophils in 14-day-old pigs decreased by 35.3% compared to one-week-old piglets but remained higher by 83.3% compared to daily piglets. The decrease in granulocytic leukocytes in the peripheral blood of animals may be associated with their migration into tissues where they form a protective barrier of the organism, as well as with their physiological apoptosis, which occurs between 14–23 days after the granulocytes enter the bloodstream[24, 25].

[24]Lerch F., Vötterl J. C., Schwartz-Zimmermann H. E., Sassu E. L., Schwarz L., Renzhammer R., Bünger M., Sharma S., Koger S., Sener-Aydemir A., Quijada N. M., Selberherr E., Kummer S., Berthiller F., U Metzler-Zebeli B. Exposure to plant-oriented microbiome altered jejunal and colonic innate immune response and barrier function more

The decrease in neutrophil fractions in 14-day-old piglets may be related to the migration of cells with cytotoxic properties from the bloodstream.

Before weaning from the sow, on the 23rd day of life, the number of lymphocytes in piglets' blood was at the same level as during the precolostral period, but compared to two-week-old animals, their content decreased by 31.7%. The fraction of neutrophils on the 23rd day, on the other hand, increased by 45.6% compared to 14-day-old piglets, but still remained significantly lower by 12.6% ($p \leq 0.05$) compared to the precolostral period. The number of monocytes and eosinophils increased by 4.6 and 7 times, respectively, by the end of the study period ($p \leq 0.01$) compared to piglets before colostrum intake.

Therefore, the postnatal period in piglets was characterized by a low absolute number of leukocytes immediately after birth, with a predominance of polymorphonuclear cells and a significant increase in their quantity until the 23rd day of life, accompanied by a redistribution of fractions towards an increase in lymphocytes, eosinophils, and monocytes and a decrease in segmented neutrophils. This may be associated with the development of the immune defense system in piglets during postnatal ontogenesis[26, 27].

The changes in leukocyte number in the first hours and days of piglets' life are influenced by colostrum intake[28, 29, 30]. These fluctuations were reflected in a sharp increase in white blood cells in the first hours after birth due to an increase in the absolute number of neutrophils, monocytes, and lymphocytes, respectively.

It is known that the number of immune cells in sow colostrum decreases during the first day of lactation compared to the first portions of colostrum,

---

strongly in suckling than in weaned piglets. *Journal of animal science*. 2022. Vol. 100, No. 11. P. skac310.

[25]Qi M., Tan B., Wang J., Liao S., Li J., Cui Z., Shao Y., Ji P., Yin Y. Postnatal growth retardation is associated with deteriorated intestinal mucosal barrier function using a porcine model. *Journal of cellular physiology*. 2021. Vol. 236, No. 4. P. 2631–2648.

[26]Annacker O., Pimenta-Araujo R., Burlen-Defranoux O., Bandeira A. On the ontogeny and physiology of regulatory T cells. *Immunological reviews*. 2001. Vol. 182. P. 5–17.

[27] Єфімов В. Г., Ракитянський В. М. Показники клітинного імунітету поросят на дорощуванні за впливу гумату натрію, бурштинової кислоти і мікроелементів. *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С. З. Ґжицького*. 2015. Т. 17, № 3. С. 32–37.

[28] Кокарєв А. В., Масюк Д. М. Формування клітинних механізмів імунного захисту у поросят за дії препарату «Імунолак». *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С. З. Ґжицького. Серія «Ветеринарні науки»*. 2017. Т. 19. № 77. С. 214–219.

[29]De Carvalho R. H., Callegari M. A., Dias C. P., Kirwan S., da Costa M. C. R., da Silva C. A. Euglena gracilis β-Glucans (1,3): Enriching Colostrum of Sow for Enhanced Piglet Immunity. *Animals: an open access journal from MDPI*. 2023. Vol. 13, No. 22. P. 3490.

[30]Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677.

while the number of these cells in the blood of newborn piglets increases[31]. This indicates the ability of leukocytes to migrate from the mother to the newborn piglets, crossing the intestinal barrier of the latter[32]. In this case, the content of maternal leukocytes in peripheral blood can take a range 25% of their total quantity, mainly due to an increase in the lymphocyte fraction. These features are due to the ability of immune cells to migrate through the body, as a result of which they enter the colostrum from the mother's blood and then enter the newborn's body, providing immune protection[33].

It should be noted that only maternal immune cells can enter the piglets' body through colostrum and participate in biological reactions there[34].

Another factor that can contribute to changes in leukocyte counts in the blood of newborn piglets is the well-known phenomenon of redistribution of stored leukocytes in respect with the background of developing alimentary leukocytosis[35, 36].

The three-time administration of the «EHCW» preparation to sows in the second half of pregnancy contributed to an increase in the number of erythrocytes, hematocrit, and hemoglobin level in piglets during the first week of life. Thus, in piglets obtained from sows in the experimental group, the hemoglobin level, hematocrit, and number of erythrocytes were on average significantly higher by 15.74%, 14.71%, and 10.13% ($p \leq 0.05$), respectively, compared to the indicators in animals of the control group, before and four hours after colostrum intake.

On the 3rd and 7th day of life, the hemoglobin level in the blood of experimental animals was significantly higher by 17.05% and 17.80% ($p \leq 0.05$), respectively, compared to the control piglets. At the same time, the

---

[31] Masiuk, D., Nedzvetsky, V., & Kokariev, A. (2023). Features of the functioning of the natural defense mechanisms of piglets under the influence of immunotropic substances. *Scientific Messenger of LNU of Veterinary Medicine and Biotechnologies. Series : Veterinary Sciences*, 25(112), 181–192.

[32] Кокарєв А. В. Формування фагоцитарної ланки імунітету поросят у ранньому постнатальному онтогенезі та її корекція препаратом «Імунолак» у ланцюзі мати-плід-новонароджений. *Збірник наукових праць Харківської державної зооветеринарної академії «Проблеми зооінженерії та ветеринарної медицини»*. Харків, 2015. Вип. 31, Ч. 2. С. 89–94.

[33] Forner R., Bombassaro G., Bellaver F. V., Maciag S., Fonseca F. N., Gava D., Lopes L., Marques M. G., Bastos A. P. Distribution difference of colostrum-derived B and T cells subsets in gilts and sows. *PloS one*. 2021. Vol. 16, No. 5. P. e0249366.

[34] Forner R., Bombassaro G., Bellaver F. V., Maciag S., Fonseca F. N., Gava D., Lopes L., Marques M. G., Bastos A. P. Distribution difference of colostrum-derived B and T cells subsets in gilts and sows. *PloS one*. 2021. Vol. 16, No. 5. P. e0249366.

[35] Madsen P. A., Etheve S., Heegaard P. M. H., Skovgaard K., Mary A. L., Litta G., Lauridsen C. Influence of vitamin D metabolites on vitamin D status, immunity and gut health of piglets. *Veterinary immunology and immunopathology*. 2023. Vol. 257. P. 110557.

[36] Wang J., Li Y., Yuan H., Shi S., Zhang L., Yang G., Pang W., Gao L., Cai C., Chu G. Effects of Alginic Acid on the Porcine Granulosa Cells and Maturation of Porcine Oocytes. *Molecular nutrition & food research*. 2023. Vol. 67, No. 22. P. e2300130. DOI: 10.1002/mnfr.202300130

number of erythrocytes in the blood of experimental piglets exceeded that of the control animals by 10.03% and 9.24% ($p \leq 0.05$), and the hematocrit index was higher by 13.12% and 11.01%, respectively. No significant differences between these indicators were found in the subsequent period.

The comparative analysis of erythrocyte indices in the groups of experimental and control animals did not reveal any statistically significant differences caused by the consumption of the experimental preparation by the experimental animals.

On the other hand, the consumption of the preparation by pregnant sows led to an increase in the total number of leukocytes in the piglets born from them by more than 50% during the first week of life. Prior to colostrum intake, the absolute number of leukocytes in the piglets of the experimental group exceeded the values of the control animals by 50.6% ($p \leq 0.05$), and after 4 hours of suckling, this difference increased to 68.7% ($p \leq 0.05$). In 7-day-old piglets of the experimental group, the number of leukocytes was higher than that of the control group by 51.2% ($p \leq 0.05$). In the subsequent early postnatal ontogenesis, the difference between the groups did not exceed 6.1%, which, given the large variability within the group, is not significant for this indicator.

The increase in the absolute number of different leukocyte fractions in the blood of the experimental group piglets was observed as a result of the action of the preparation (Table 4).

In the blood of experimental animals after consuming colostrum, the fractions of neutrophils and lymphocytes statistically prevailed by 1.58 times ($p \leq 0.05$) and 2.11 times ($p \leq 0.01$), respectively, compared to the pigs in the control group. At the same time, there was a tendency for an increase in monocytes by 66.67% in the experimental piglets.

No differences were found between three-day-old piglet groups in terms of neutrophil levels. However, the experimental group of animals exceeded the control group by more than 30% ($p \leq 0.05$) in terms of mononuclear cell content.

By the end of the first week of life, the difference between the groups in terms of lymphocyte content likely increased to 80.86% ($p \leq 0.01$), for monocyte count – up to 88.24%, and for neutrophils – up to 29.54%. In the subsequent 16 days of the study, no significant differences were found between the groups.

Table 4

Number of different forms of leukocytes in pig blood during the early period of postnatal ontogenesis and their correction with the «EHCW» preparation (M±m, cells/μL, n=10)

Age		B	E	Neutrophils		L	M
				R	S		
0 hours	control	0	0,02±0,01	0,06±0,01	1,86±0,14	1,36±0,17	0,03±0,01
	«EHCW»	0	0,04±0,01	0,08±0,01	2,56±0,36	2,27±0,29	0,05±0,02
4 hours	control	0	0,05±0,03	0,18±0,04	4,52±0,37	1,86±0,33	0,09±0,04
	«EHCW»	0	0,11±0,06	0,18±0,05	6,95±1,04*	3,92±0,33**	0,15±0,05
3 days	control	0	0,08±0,02	0,08±0,03	3,04±0,19	3,47±0,29	0,11±0,02
	«EHCW»	0	0,07±0,02	0,15±0,08	2,70±0,53	4,57±0,27*	0,15±0,04
7 days	control	0	0,17±0,04	0,21±0,02	4,74±0,61	4,44±0,57	0,17±0,04
	«EHCW»	0	0,22±0,08	0,20±0,06	5,94±0,63	8,03±0,74**	0,32±0,13
14 days	control	0	0,09±0,04	0,14±0,03	2,97±0,42	5,40±0,64	0,25±0,09
	«EHCW»	0	0,11±0,02	0,08±0,03	2,99±0,17	4,94±0,43	0,18±0,06
23 days	control	0	0,64±0,15	0,27±0,08	7,25±0,56	6,15±0,54	0,46±0,20
	«EHCW»	0	0,40±0,09	0,30±0,07	7,33±0,68	6,97±0,34	0,38±0,10

Note: * – p≤0.05; ** – p≤0.01 compared to control.

Thus, the administration of the «EHCW» preparation to pregnant sows in the second half of gestation ensures a significantly higher level of leukocytes in the resulting piglets during the first week of life, mainly due to an increase in the fraction of mononuclear leukocytes and neutrophils. These data indicate the immunomodulatory effect of the preparation on the organism of newborn piglets[37]. This effect can initiate the enhancement of their cellular immune defense[38].

[37]Le Huërou-Luron I., Bouzerzour K., Ferret-Bernard S., Ménard O., Le Normand L., Perrier C., Le Bourgot C., Jardin J., Bourlieu C., Carton T., Le Ruyet P., Cuinet I., Bonhomme C., Dupont D. A mixture of milk and vegetable lipids in infant formula changes gut digestion, mucosal immunity and microbiota composition in neonatal piglets. *European journal of nutrition.* 2018. Vol. 57, No. 2. P. 463–476.

[38]Lerch F., Vötterl J. C., Schwartz-Zimmermann H. E., Sassu E. L., Schwarz L., Renzhammer R., Bünger M., Sharma S., Koger S., Sener-Aydemir A., Quijada N. M., Selberherr E., Kummer S., Berthiller F., U Metzler-Zebeli B. Exposure to plant-oriented microbiome altered jejunal and colonic innate immune response and barrier function more strongly in suckling than in weaned piglets. *Journal of animal science*. 2022. Vol. 100, No. 11. P. skac310.

The analysis of the obtained results allows us to conclude that the early neonatal period of ontogenesis was characterized by a sufficiently high absolute number of erythrocytes, hematocrit level, and hemoglobin in piglets after birth, followed by a decrease in these indicators during the first seven days of life by 14.18% ($p \leq 0.05$), 20.8% ($p \leq 0.05$), and 17.6% ($p \leq 0.05$), respectively, and an increase in these indicators in the subsequent 16 days of life compared to the indicators in animals at the time of birth.

In newborn piglets, innate immunity during the early period of postnatal ontogenesis was characterized by a low absolute number of leukocytes at the time of birth, with neutrophils prevailing, and an increase in leukocyte count by 2.02 times ($p \leq 0.01$) in the first hours of life due to a 21.9% increase ($p \leq 0.01$) in the fraction of neutrophils and a simultaneous 33.0% decrease ($p \leq 0.01$) in lymphocytes. Subsequently, the leukocyte number gradually increased to 2.9 and 4.45 times ($p \leq 0.001$) on the 7th and 23rd days of life, respectively, compared to the indicators at birth. These changes were accompanied by a decrease in the number of neutrophils and an increase in the number of lymphocytes and monocytes on the 14th day of life by 1.4 times ($p \leq 0.001$), 1.5 times ($p \leq 0.01$), and 3.7 times ($p \leq 0.05$), respectively, compared to the indicators at birth.

These characteristics are determined by the fact that piglets are born at an earlier stage of intrauterine development than other mammals[39]. Against the background of the biological peculiarities of the structure of the placenta in pigs and the complex action of stressors on piglets immediately after birth, the formation of physiological immunodeficiency occurs, which is the first of the three main critical periods of postnatal ontogenesis[40].

Correction of pregnant sows with the «EHCW» preparation contributed to an increase in the level of hemoglobin by 16.6% ($p \leq 0.05$), the number of erythrocytes by 9.9% ($p \leq 0.05$), and leukocytes by more than 45.8% ($p \leq 0.05$) in piglets during the first week of life. This was achieved through an increase in the fractions of lymphocytes and segmented neutrophils by an average of 1.73 times ($p \leq 0.05$- $p \leq 0.01$) and 1.27 times ($p \leq 0.05$), respectively, compared to the indicators in the animals of the control group.

---

[39]Dong L., Zhong X., Zhang L., Kong L., Kong Y., Kou T., Wang T. Impaired intestinal mucosal immunity is associated with the imbalance of T lymphocyte sub-populations in intrauterine growth-restricted neonatal piglets. *Immunobiology*. 2015. Vol. 220, No. 6. P. 775–781.

[40]Martinez C. A., Rubér M., Rodriguez-Martinez H., Alvarez-Rodriguez M. Pig Pregnancies after Transfer of Allogeneic Embryos Show a Dysregulated Endometrial/Placental Cytokine Balance: A Novel Clue for Embryo Death? *Biomolecules*. 2020. Vol. 10, No. 4. P. 554.

2. Cellular immunity

The results of present study on the development of cellular immunity indicate that in the first hours after birth, pig blood containes a high level of active phagocytes, which gradually decrease over time, while their aggressiveness and digestive capacity, on the contrary, increased. The application the «EHCW» preparation in sows induces an increase in the content of active phagocytes and a simultaneous enhancement of their aggressiveness as well as stimulates digestive capacity which were observed in the piglets born from them.

The measuring of the dynamics of active phagocyte content in pig blood during the early postnatal period make found that the index of phagocytic activity (PhA) had the highest value on the first day of life and gradually decreased over time. Prior to consuming colostrum and in the first hours after suckling, it ranged around 77% (Figure 1), which during the period before colostrum intake and in the first hours of life forms the main immunological barrier of newborns[41].

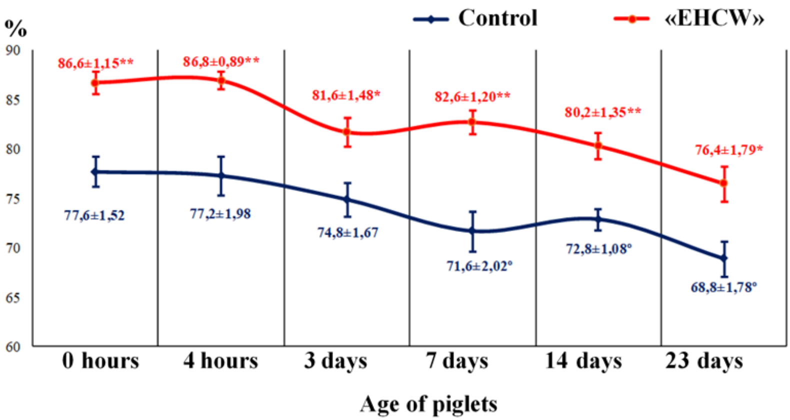

Fig. 1. Dynamics of phagocytic activity of leukocytes in the blood of newborn piglets at different stages of the neonatal period and its correction with the «EHCW» preparation (M±m, n = 10)

Note: 1. 0–p≤0.05; 00–p≤0.01 in relation to piglets before colostrum intake; 2. *–p≤0.05; **–p≤0.01 in relation to the control.

[41]Farmer C., Edwards S. A. Review: Improving the performance of neonatal piglets. *Animal: an international journal of animal bioscience.* 2022. Vol. 16, Suppl 2. P. 100350.

Subsequently, on days 3 and 7, this index decreased to 74% and 71%, respectively, compared to the values in animals before colostrum intake.

During this time, piglets experience the development of anemia, which is caused by an iron deficiency. Iron is a critically important element for the functional activity of immune cell enzymes that generate oxides and peroxides, which overall affects the functional activity of immune cells and directly influences the level of activity of neutrophils with activated oxygen-dependent bactericidal mechanisms. In the absence of iron, the activity of protein kinase C decreases due to its trantbocation in T-lymphocytes[42, 43].

Throughout the second week of life, PhA in the blood of control group piglets insignificantly increased to almost 73%, and by the end of the third week, it decreased again to 69% compared to the values in animals at 7 days of age. In our opinion, the tendency of a decrease in the number of active phagocytes in piglets on days 3 and 7 is due to the formation of colostral defense mechanisms, and by the end of the third week, the formation of their own immunobiological factors.

Thus, the early postnatal period of piglet ontogenesis was characterized by a high level of phagocytic activity of leukocytes in newborns on the first day of life, which gradually decreased during the first and third weeks. This decrease is associated with low activity of humoral defense factors during this time. Subsequently, during passive immunization, newborns receive physiological immunity that has formed in the mother's body under the influence of antigenic pressing. This allows activating defense mechanisms in animals during the initial stage of postnatal adaptation[44].

The study of PhA in the blood of neonatal piglets after correction with the «EHCW» preparation showed an increase in this parameter in the experimental animals throughout the lactation period. The difference between the groups was almost 11% ($p \leq 0.01$) immediately after birth. By day 3, it slightly decreased to 8.3% ($p \leq 0.05$). Contrary, it increased to 13.3% ($p \leq 0.01$) by day 7. By the end of the second weeck and at the beginning of the fourth week of life, phagocytic activity in the experimental piglets significantly exceeded that in the control animals by an average of 9.5%. This may be attributed to the increased influx of opsonins from the

[42]Bréa D., Soler L., Fleurot I., Melo S., Chevaleyre C., Berri M., Labas V., Teixeira-Gomes A. P., Pujo J., Cenac N., Bähr A., Klymiuk N., Guillon A., Si-Tahar M., Caballero I. Intrinsic alterations in peripheral neutrophils from cystic fibrosis newborn piglets. *Journal of cystic fibrosis: official journal of the European Cystic Fibrosis Society*. 2020. Vol. 19, No. 5. P. 830–836.

[43]Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677.

[44]Kulkarni D. H., Newberry R. D. Intestinal Macromolecular Transport Supporting Adaptive Immunity. *Cellular and molecular gastroenterology and hepatology*. 2019. Vol. 7, No. 4. P. 729–737.

mother's colostrum and increased expression of plasma membrane receptors on phagocytic cells, which possess adhesion properties towards antigenic structures without the involvement of opsonins.

Therefore, the use of the preparation contributed to an increase in the level of PhA in the blood of piglets, which may indicate an enhancement of their defense level against the development of age-related immunodeficiency states during the lactation period.

The study of the aggressiveness and digestive capacity of activated phagocytes in the blood of piglets during the early postnatal period indicates a gradual increase in leukocyte aggressiveness throughout the entire research period. There was a slight increase in the digestive capacity of leukocytes during the first week after birth. The level of digestive capacity sharply increased during the second week of life and returned to the level of 7-day-old piglets by 23th day after birth.

The dynamics of phagocytic cell number and the state of their digestive capacity had a progressive character (Table 5).

Table 5

Phagocytic Cell Quantity (PhCQ) and Digestive Capacity Index (PHA) in the blood of piglets during the early postnatal ontogenesis and their correction with the «EHCW»preparation (M±m, Un., n = 10)

Age	PhCQ		PHA	
	control	«EHCW»	Контроль	Дослід
0 hours	2,12±0,21	2,30±0,18	1,27±0,17	1,40±0,19
4 hours	2,28±0,10	2,60±0,17	1,34±0,11	1,35±0,23
3 days	2,62±0,20	3,08±0,33	1,39±0,19	1,56±0,15
7 days	2,68±0,20	3,28±0,16*	1,47±0,20	1,67±0,16
14 days	2,94±0,15^{0}	3,18±0,14	1,86±0,15^{0}	2,23±0,16
23 days	3,10±0,19^{00}	3,16±0,18	1,53±0,19	2,20±0,21*

Note: 0 – p ≤ 0.05; 00 – p ≤ 0.01 compared to piglets before colostrum consumption; 2. * – p ≤ 0.05; ** – p ≤ 0.01 compared to control.

The index of phagocyte aggressiveness was at a level of 2.2 Un in the piglet blood on the first day of life. Within three days, this indicator has increased by almost 20%, and by the end of the first week, it remained at this level.

Over the next 14 days, the level of leukocyte aggressiveness was gradually increased, as evidenced by an increase in PhCQ by 9.7% and 15.7% on the 14th and 23rd day of life of neonatal piglets, respectively, compared to the 7th day of life. Comprehensive level of this indicator between piglets before colostrum consumption and on the 14th and 23rd day of life, an increase of 38.7% and 46.2% was observed, respectively, compared to the pre-colostrum consumption values. The increase in PhCQ in piglets during the lactation period indicates an enhancement of their phagocytic capacity. Since phagocytosis is a one of engine of cell movement, it can be assumed that the migratory ability of phagocytic cells also increases over time, providing a significantly higher level of cellular immunity[45, 46].

As mentioned above, the dynamics of the digestive capacity of active leukocytes in the blood of newborn piglets during the first two weeks showed an ascending pattern. Immediately after birth, before colostrum consumption, PhA had the lowest value throughout the experimental period, at 1.27 Un. Since the value is greater than 1, it indicates that the process of phagocytosis is complete.

During the first week of life, PhA in the control piglets gradually increased. On the 7th day after birth, this indicator was 1.67 Un., which was 19.3% higher than its initial level. Over the next 7 days, there was a sharp increase in the digestive capacity of active leukocytes. During the same period, PhA reached its maximum value throughout the experimental period, which was 26.5% higher than the previous value and 46.5% higher compared to newborn animals. In the following 7 days of life, PhA decreased to 1.53 Un., which was 17.7% lower than the value at 14th days.

Thus, the early postnatal period of piglet development was characterized by an ascending dynamic of aggressiveness and the completeness of the digestive capacity of active leukocytes throughout the lactation period, with a decrease in the level of the latter at the beginning of the fourth week of life.

When investigating the immunomodulatory effects of the «EHCW» drug on phagocytic activity (PhA) and immunezyme activity (IA) in the blood of newborn piglets, it was found that both indicators in the experimental group of piglets exceeded the values of the control group by 8.5% and 10.2%, respectively, before colostrum intake. Four hours after colostrum suckling, PhA in the experimental piglets increased by 14.0% compared to the control animals. At the same time, IA in the experimental animals decreased to the same level as the control. On the third day of life, the PhA and IA values in the blood of the experimental group piglets were higher than those in the control animals by 17.6% and 12.2%, respectively.

---

[45] Aegerter H., Lambrecht B. N., Jakubzick C. V. Biology of lung macrophages in health and disease. *Immunity*. 2022. Vol. 55, No. 9. P. 1564–1580.

[46] Bonilla M.C., Fingerhut L., Alfonso-Castro A., Mergani A., Schwennen C., von Köckritz-Blickwede M., de Buhr N. How Long Does a Neutrophil Live?-The Effect of 24 h Whole Blood Storage on Neutrophil Functions in Pigs. *Biomedicines*. 2020. Vol. 8, No. 8. P. 278.

By the end of the first week, the difference between the groups in terms of FA showed significant changes and amounted to 22.4% ($p \leq 0.05$). The level of IA at this time was higher in the experimental animals by 13.6% compared to the control group. By the end of the second week of life, the difference between the groups of piglets in these indicators decreased by 8.2% and almost 20%, respectively. On the 23rd day of life, the phagocytic count between the experimental and control animals was almost the same, while IA in the experimental group of animals was significantly higher by 43.8% ($p \leq 0.05$) compared to the indicators in the control group animals.

Thus, the immune correction of sows with the «EHCW» drug application in the second half of pregnancy leads to an increase in the aggressiveness of phagocytic leukocytes during the first week of life and enhances their digestive capacity from the second week after birth. This, in turn, enhances the level of protection of newborns, especially during the critical period, which arises due to the expenditure and catabolism of colostral immune factors in the absence of a fully formed immune system[47, 48].

During the early postnatal period, piglets had a relatively high level of nitrotetrazolium blue test (NTB). This index had high values in animals at 1 and 14 days of age. A slight decrease was observed in sows on the 7th day after birth, and a sharp decline was noted at the beginning of the 4th week of life.

The NTB level was established 18.8% in newborn piglets of the control group, before colostrum intake (Figure 2). This index was increased to 19.8% four hours after suckling start in compare with the levels in animals before colostrum intake.

On the 3rd and 7th day, the NTB decreased by 1.21 and 1.25 times, respectively, compared to the same value in piglets on the first day of life. During the second week, NTB in piglets increased by 1.22 times compared to piglets at 7 days of age, and by the end of the third week, it sharply decreased by almost 1.46 times compared to the value at 14 days of age.

Thus, the formation of the phagocytic branch of immunity in piglets during the early postnatal ontogenesis period was characterized by a high content of microphages with an active oxygen-dependent metabolism mechanism at the daily and two-week ages, and a decrease in their activity in the second half of the first and the beginning of the fourth week after birth.

---

[47]Cheng Z., Zhou S. T., Zhang X. H., Fu Q., Yang Y., Ji W. B., Liu H. G. Effects of early intermittent maternal separation on behavior, physiological, and growth performance in piglets. *Journal of animal science*. 2023. Vol. 101. P. skad122.

[48] Єфімов В. Г., Ракитянський В. М. Показники клітинного імунітету поросят на дорощуванні за впливу гумату натрію, бурштинової кислоти і мікроелементів. *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С. З. Ґжицького.* 2015. Т. 17, № 3. С. 32–37.

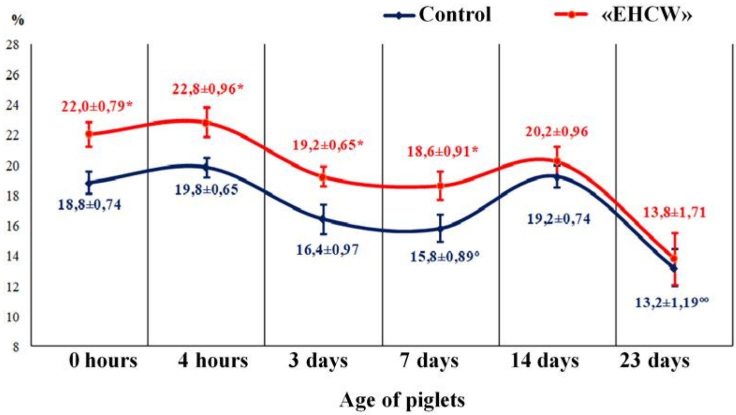

Fig. 2. NTB indicator in the blood of piglets during the early postnatal ontogenesis period and its correction with the «EHCW» drug (M±m, %, n=10)

Note: 1. 0–p≤0.05; 00–p≤0.01 compared to piglets before colostrum intake; 2. *–p≤0.05; **–p≤0.01 compared to the control.

The decrease in enhanced digestive capacity of neutrophilic leukocytes alongside a decrease in its quantity may be due to a compensatory response of the pig's organism and associated with the stimulation of lysosomal enzymatic systems in neutrophils. This occurs due to an increase in antigenic pressing in the pig's body, which activates phagocytes[49, 50].

Under the action of the «EHCW» drug, an increase in NTB levels was observed in newborn piglets during the first week of life. It should be noted that in the experimental animals, before colostrum intake, the NTB values significantly exceeded those of the control group by 17.1% ($p \leq 0.05$). After four hours, this difference decreased to 13.2% ($p \leq 0.05$), and by 3 and 7 days of life, it was restored to 14.6% and 15.1% ($p \leq 0.05$), respectively. Starting from day 14 after birth and onwards, no differences were found between the experimental and control groups.

[49] Bonilla M.C., Fingerhut L., Alfonso-Castro A., Mergani A., Schwennen C., von Köckritz-Blickwede M., de Buhr N. How Long Does a Neutrophil Live?-The Effect of 24 h Whole Blood Storage on Neutrophil Functions in Pigs. *Biomedicines*. 2020. Vol. 8, No. 8. P. 278.

[50] Bréa D., Soler L., Fleurot I., Melo S., Chevaleyre C., Berri M., Labas V., Teixeira-Gomes A. P., Pujo J., Cenac N., Bähr A., Klymiuk N., Guillon A., Si-Tahar M., Caballero I. Intrinsic alterations in peripheral neutrophils from cystic fibrosis newborn piglets. *Journal of cystic fibrosis: official journal of the European Cystic Fibrosis Society*. 2020. Vol. 19, No. 5. P. 830–836.

Correction of pregnant sows with the «EHCW» drug promotes an increase in the number of microphages with activated oxygen-dependent bacteriolytic activity during the first week of piglet life.

Analyzing the above information, it should be noted that the early postnatal period of piglet ontogenesis was characterized by a high content of active phagocytes in the blood of newborns, including those with an active oxygen-dependent mechanism of bacteriolytic activity, and its gradual decrease in the second half of the first week and throughout the third week of life. At the same time, the activity and aggressiveness of phagocytes increased alongside their enhanced digestive capacity.

The analysis of the immunoregulatory effect of the drug from the cell wall of lactobacilli showed that throughout the entire observation period, the PhA indicator in the blood of newborn piglets was 11.5% higher compared to the animals in the control group. The difference between the groups in terms of the PhCQ had an upward growth trend, with the peak occurring at the end of the first week of life, after which it started to decrease and disappeared at the beginning of the fourth week. At the same time, there was an increase in the NTB indicator during the first 7 days of life, on average by 17% compared to the control group.

The results of lymphocyte studies showed that before colostrum consuming, undifferentiated forms of lymphocytes predominated in the blood of piglets (Table 6), as most agranulocytes in newborn animals do not have differentiating receptors.

Table 6

Dynamics of lymphocytes content in the blood of piglets from the control group during the early postnatal period of ontogenesis (M±m, %, n=10)

Age	**Lymphocytes**			
	T-	**B-**	**NK-**	**0-**
0 hours	14,90±1,08	10,10±0,99	4,40±0,80	70,60±1,49
4 hours	26,00±2,23**	17,80±1,81**	7,60±0,78*	48,60±2,53***
3 days	29,20±2,34***	20,20±1,33***	7,10±0,94	43,50±1,32***
7 days	35,40±1,97***	21,30±0,95***	9,40±0,65**	33,90±1,54***
14 days	30,80±1,53***	19,50±0,94***	8,90±0,94**	40,80±1,39***
23 days	41,30±1,05***	22,30±1,05***	8,70±0,96**	27,70±1,97***

Note: *–p≤0.05; **–p≤0.01; ***–p≤0.001 compared to piglets at 0 hours.

Four hours after colostrum intake, there was a redistribution among lymphocytes. During this time, the relative proportion of 0-lymphocytes likely decreased by 31.2% ($p \leq 0.001$), while the proportions of T-, B-, and NK-lymphocytes increased by 74.5% ($p \leq 0.01$), 76.2% ($p \leq 0.01$), and 72.7% ($p \leq 0.05$), respectively, compared to the values before colostrum intake.

During the first 7 days of life, the relative proportion of differentiated lymphocyte forms in the blood of piglets continued to gradually increase, while the fraction of 0-cells, conversely, decreased.

The table shows the dynamics of lymphocytes in the blood of piglets from the control group during the early postnatal period of ontogenesis

During the first 3 days of life, the number of T-, B-, and NK-lymphocytes increased by 1.9, 2.0 ($p \leq 0.001$), and 1.6 times, respectively, due to a decrease in the content of 0-lymphocytes by 38.4% compared to piglets before colostrum intake. In the following 4 days, the changes were less pronounced and characterized by an increase in the fractions of T-, B-, and NK-lymphocytes by 21.2%, 5.5%, and 32.4%, respectively, along with a simultaneous decrease in 0-lymphocytes by 22.1% compared to animals at 3 days of life.

By the end of the second week of life, there was a decrease in the relative number of T-, B-, and NK-cells by 13.0%, 8.5%, and 5.3%, respectively, along with an increase in the fraction of 0-lymphocytes by 20.4% compared to piglets at one week of age. These changes may be due to apoptosis of short-lived lymphoid cells, the number of which in circulating blood can reach 30%, or migration of cells with cytotoxic properties out of the bloodstream.

A comparative analysis of lymphocyte counts in 14-day-old piglets showed a statistically significant increase in all differentiated forms of lymphocytes by an average of 2.0 times ($p \leq 0.001$) and a decrease of 42.2% ($p \leq 0.001$) in the number of 0-lymphoid cells.

A decrease in the relative number of T-, B-, and NK-lymphocytes, along with an increase in the fraction of 0-lymphocytes, may be attributed to the apoptosis of short-lived lymphoid cells, the number of which in the circulating blood can reach 30%. Such changes occur due to the action of antigen-presenting cells in lymphoid follicles, where they participate in the differentiation of naïve lymphocytes into effector short-lived cells. The majority of these cells undergo apoptosis 4-7 days after differentiation, while others transform into memory immune cells that provide a faster and more intense response to antigen rechallenge[51, 52].

Simultaneously, a decrease in the number of differentiated lymphocytes alongside the increase in 0-cells can be caused by the previous depletion

[51]Pietrasina O., Miller J., Rząsa A. Differences in the relative counts of peripheral blood lymphocyte subsets in various age groups of pigs. *Can J Vet Res*. 2020 Vol. 84, No. 1. P. 52–59.

[52]Sinkora M., Butler J. E. Progress in the use of swine in developmental immunology of B and T lymphocytes. *Developmental and comparative immunology*. 2016. Vol. 58. P. 1–17.

of iron reserves in piglets' bodies, as the activity of ribonucleotide reductase enzyme, responsible for DNA synthesis and cell replication, is iron-dependent. Iron regulates the production of interleukin-2, which is one of the main cytokine, but not the only this cytokine involved in the proliferation and differentiation of various T-, B-, and NK-lymphocytes populations[53, 54].

On the 23rd day of life, the highest number of differentiated lymphocyte forms was observed in the peripheral blood of piglets, accounting for 72.3% of the total lymphocyte pool, among which 57% were T-lymphocytes, 31% were B-lymphocytes, and the remaining 12% were NK-lymphocytes. It should be noted that at this age, the percentage of T- and B-lymphocytes was the highest throughout the study period, surpassing the values of suckling piglets by 2.8 and 2.2 times, respectively ($p \leq 0.001$), and the values of piglets at the age of 2 weeks by 34.1% and 14.4%, respectively, which is associated with the development of the piglets' immune system.

The NK-lymphocyte fraction goes to its maximum content peak on the 7th day of piglets' life, after which it slightly decreased by 7.5%, but remained significantly higher by 2.0 times ($p \leq 0.001$) compared to animals in the non-lactation period and fluctuated at this level until the end of the study. These changes are likely associated with the activating action of immuno-biological substances present in the mother's colostrum, which contribute to the formation of the piglets' immune system.

Correlation analysis of the relationships between age-related changes in different lymphocyte classes indicates that the relative content of T-, B-, and NK-lymphocytes negatively correlates with the fraction of 0-cells. The highest degree of correlation is observed between T- and 0-lymphocytes ($r = -0.99$), followed by a slightly lower dependency between B- and 0-lymphocytes ($r = -0.98$), and NK- and 0-lymphocytes ($r = -0.92$).

The main population among differentiated lymphocyte forms is comprised of T-cells. Analysis of the distribution of this population based on the density of receptors on the cell membrane surface during the first 23 days of piglets' life has shown that the majority of these cells had low receptor density, resulting in their age dynamics reflecting the overall changes in the total number of T-lymphocytes (Table 7).

---

[53]Ray A., Bhati T., Arora R., Rastogi S. Progesterone-mediated immunoregulation of cytokine signaling by miRNA-133a and 101-3p in Chlamydia trachomatis-associated recurrent spontaneous abortion. *Molecular immunology*. 2023. Vol. 164. P. 47–57.

[54]Masiuk D. M., Kokariev A. V., Bal R., Nedzvetsky V. S. The isotonic protein mixture suppresses Porcine Epidemic Diarrhea Virus excretion and initiates intestinal defensive response. *Theoretical and Applied Veterinary Medicine*. 2022. Vol. 10, No. 2. P. 23–28.

Table 7

Dynamics of T-lymphocytes with different membrane receptor density in the blood of control group piglets during the early period of postnatal ontogenesis (M±m, %, n=10)

Age	T-lymphocytes		
	LDMR	MDMR	HDMR
0 hours	13,80±0,88	1,10±0,21	0
4 hours	19,70±1,49**	4,90±0,65***	1,40±0,21***
3 days	23,20±1,29***	4,80±0,91**	1,20±0,29**
7 days	26,80±1,04***	6,90±0,87***	1,70±0,14***
14 days	22,30±0,78***	7,40±0,78***	1,10±0,21***
23 days	29,60±1,02***	9,30±0,86***	2,40±0,33***

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to 0-hour-old piglets.

Before suckling colostrum, their number in piglets' blood accounted for 92.62%, and within four hours after birth, their count decreased to 75.77% relative to the total number of T-cells, remaining at this level with fluctuations within 4% until the 23rd day of life.

Analysis of the age dynamics of T-lymphocyte content with low density of membrane receptors (LDMR) showed that their quantity gradually increased throughout the entire neonatal period with a slight decrease on the 14th day of life. Just a few hours after suckling colostrum, the number of these cells increased by 42.75% ($p \leq 0.01$) compared to the previous value. By the 3rd day after birth, this difference grew to 68.12% ($p \leq 0.001$), and by the end of the first week of life, their quantity exceeded the value of non-lactating piglets by almost 2 times ($p \leq 0.001$).

Throughout the initial two weeks of life, there was a slight decrease in the quantity of T-lymphocytes with LDMR by 16.79% compared to piglets at 7 days of age. However, their quantity remained significantly higher by 61.59% ($p \leq 0.001$) compared to non-lactating piglets.

The second largest T-lymphocyte population in the blood of piglets was the population with middle density of membrane receptor (MDMR), while the smallest percentage was represented by cells with high receptor density on the plasma membrane. Prior to colostrum intake, the quantity of cells with MDMR accounted for 7.38%, and lymphocytes with high density of membrane receptors (HDMR) were completely absent.

A few hours after suckling colostrum, the percentage of T-cells with MDMR in the blood of newborn piglets increased by 4.46 times ($p \leq 0.001$), and T-lymphocytes with HRMD appeared, comprising 5.39% of the total

T-cell count compared to the values at the time of birth. The quantity of these lymphocyte subsets remained at this level until the 3rd day of age.

Subsequently, at 7, 14, and 23 days of life, there was a gradual increase in the quantity of T-lymphocytes with MDMR in the blood of piglets by 40.81%, 51.02%, and 89.79% respectively, compared to their quantity in 3-day-old pigs, and by 6.27, 6.73, and 8.46 times ($p \leq 0.001$) respectively, compared to the values at the time of birth.

The quantity of T-cells with HDMR in piglets at 7 days of age increased by 41.67% compared to 3-day-old animals, and by the end of the second week of life, it decreased by 35.29% compared to pigs at one week of age.

An increase in the quantity of T-lymphocytes with HDMR was observed in the blood of piglets on the 23rd day, reaching 2.18 times compared to two-week-old pigs.

Early postnatal period in piglets was characterized by a predominance of low-density receptor T-lymphocytes in the blood, with a reduced number of cells possessing high-density membrane receptors. Subsequently, the neonatal period in pigs was accompanied by a gradual increase in the T-lymphocytes population that consist the cells with low and medium membrane receptor densities, until the twenty-third day of life. There was a slight decrease in the number of low-affinity cells at two weeks of age, as well as a parabolic change in the number of high-affinity T-cells, with peaks on the seventh and twenty-third day of life and a decrease at the end of the second week after birth[55].

Studies on the ratio of B-lymphocytes with different membrane receptor densities indicate that, prior to colostrum intake, the majority of these cells in piglets were lymphocytes with low receptor density on the plasma membrane (Table 8). Their quantity accounted for 97.03% of the total content. The remaining nearly 3% consisted of cells with medium receptor concentrations, while high-affinity B-lymphocytes were not detected in piglets at the time of birth.

Four hours after birth, the number of B-lymphocytes in piglets' blood increased due to an increase in cells with low and medium membrane receptor densities by 1.60 times ($p \leq 0.01$) and 5.67 times ($p \leq 0.05$), respectively. At the same time, cells with high receptor densities were detected, accounting for 2.25% of the total pool of B-cells.

[55]Pietrasina O., Miller J., Rząsa A. Differences in the relative counts of peripheral blood lymphocyte subsets in various age groups of pigs. *Can J Vet Res*. 2020 Vol. 84, No. 1. P. 52–59.

Table 8

B-lymphocytes with different membrane receptor densities in the blood of piglets from the control group during the early postnatal ontogenesis period (M±m, %, n = 10)

Age	B-lymphocytes		
	LDMR	MDMR	HDMR
0 hours	9,80±0,88	0,30±0,14	0
4 hours	15,70±1,27**	1,70±0,45*	0,40±0,21*
3 days	17,70±0,89***	1,90±0,27***	0,60±0,21**
7 days	18,60±0,93***	1,90±0,21***	0,80±0,22
14 days	17,50±0,59***	1,60±0,21***	0,40±0,21**
23 days	19,10±0,72***	2,20±0,29***	1,00±0,31

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to piglets at 0 hours of age

The increase in B-lymphocytes in pigs at 3 days of age occurred due to an elevation of all forms of these cells in the blood. Lymphocytes with LDMR increased by 80.61% ($p \leq 0.001$), with MDMR by 6.33 times ($p \leq 0.001$) compared to the values at the time of birth, and with high-density receptors by 50% compared to the values in the first few hours of life. Changes in the number of B-cells in one-week-old pigs were due to an increase in two fractions of B-lymphocytes – with HDMR and LDMR, by 33.33% and 5.08%, respectively, compared to the previous value, while the level of cells with MDMR remained unchanged.

A decrease in the number of B-lymphocytes observed in 14-day-old piglets occurred due to a simultaneous decrease in cells with low, medium, and high receptor avidity by 5.91%, 15.79%, and 50.00%, respectively, compared to the values in one-week-old pigs. However, despite this decrease, the number of low- and medium-affinity B-lymphocytes remained 1.79 and 5.33 times higher ($p \leq 0.001$), respectively, compared to the values at the time of birth, while the content of high-affinity cells reached the level observed in the first few hours of life.

Furthermore, the increase in B-lymphocytes in twenty-three-day-old piglets occurred due to an elevation of cells with LDMR by 9.14%, MDMR by 37.50%, and HDMR by 2.5 times compared to the values in two-week-old piglets.

Thus, the neonatal period of piglet development was characterized by a predominance of low-affinity B-lymphocytes in the blood before colostrum intake, with the absence of high-affinity cells. Postnatal changes in the quantity of antibody-producing cell precursors in piglets at all stages of the study were influenced by cells of all three receptor avidity types, and by the end of the first week of life, only B-lymphocytes with high and low membrane receptor densities were responsible for these changes.

Changes in the lymphocyte fractions of piglets during the neonatal period were characterized by redistribution of differentiated and undifferentiated cell forms in the first day of life and the appearance of cells with high density of membrane receptors in the blood of piglets a few hours after birth. This was followed by a gradual increase in the number of T- and B-lymphocytes over a period of 23 days, with a slight decrease on the 14th day due to proportional changes in cell fractions with different membrane receptor densities.

Studies on the ratio of T-lymphocyte subpopulations in the peripheral blood of piglets at different stages of early postnatal ontogenesis indicate a gradual increase in the number of theophylline-resistant forms of T-lymphocytes throughout the suckling period, with a slight decrease on the 14th day of life (Figure 3). The lowest level of theophylline-resistant T-lymphocytes was found in the blood of piglets before colostrum intake, and after 4 hours of suckling from the sow, their level increased by almost 2.4 times (p≤0.01) compared to the levels at birth.

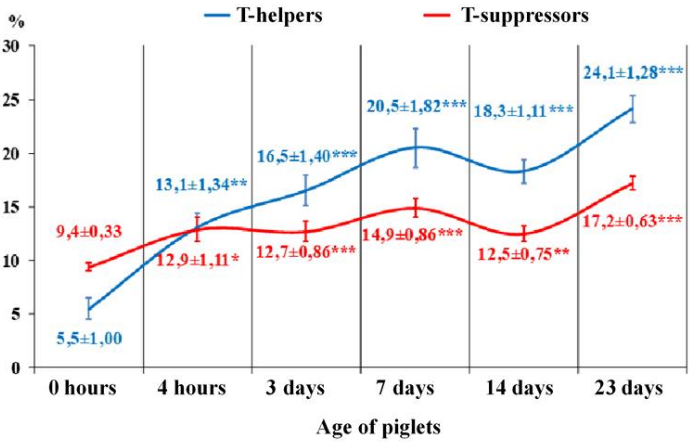

Fig. 3. The number of T-helpers cells and T-suppressors cells in the blood of piglets in the control group during the early period of postnatal ontogenesis (M±m, n=10)

Note: * – p≤0.05; ** – p≤0.01; *** – p≤0.001 compared to piglets at 0 hours of age.

On the third day, the number of T-helper cells increased by 3.0 times ($p \leq 0.001$) compared to the levels at birth and by 26.0% compared to the piglets of the first day of life. By the end of the first week, the fraction of theophylline-resistant lymphocytes increased by an additional 24.2% compared to the piglets at three days of age and was 3.7 times ($p \leq 0.001$) higher than the levels at birth. In the following 16 days, there was an increase in the number of T-helper cell forms by 17.6%, with a slight decrease of 10.7% on the 14th day compared to animals at 7 days of age.

The obtained results of the study on age-related changes in the number of theophylline-sensitive lymphocytes in piglets during the early period of postnatal ontogenesis allow us to note that their content was the lowest before colostrum intake, relative to the entire suckling period. After 4 hours of suckling from the sows, the number of theophylline-sensitive cells in the blood of piglets likely increased by 37.2% ($p \leq 0.05$) compared to the levels at birth. At this level, their quantity remained stable during the first 3 days of life, and by the end of the first week, it increased again by 15.5% compared to piglets of the first day of life after colostrum intake. Subsequently, the changes that occurred in the fraction of theophylline-sensitive lymphocytes were similar to the theophylline-resistant cells and were characterized by a slight decrease of 16.1% in 14-day-old animals and an increase of 15.4% in 23-day-old animals, compared to week-old pigs. It was found that before colostrum intake, the fraction of theophylline-sensitive T-lymphocytes predominated over the theophylline-resistant cells by 1.7 times.

After colostrum intake, a redistribution of lymphocyte populations were observed, resulting in an equal number of T-helper and T-suppressor cells in the blood of piglets on the first day of life. However, in piglets at 3 days of age, the fraction of theophylline-resistant lymphocytes exceeded the fraction of theophylline-sensitive cells by almost 30%. On days 7 and 14 of life, this difference increased to 37.6% and 46.4%, respectively, and slightly decreased to 40.1% on day 23. The study of age-related changes in theophylline-resistant and theophylline-sensitive T-lymphocytes with different receptor density on their membrane reflected the dynamics of these cells, which had a direct correlation with changes observed in the ratio of total T-lymphocytes (Table 9).

Table 9

The number of theophylline-resistant and theophylline-sensitive T-lymphocytes with different membrane receptor densities in the blood of piglets in the control group during the early period of postnatal ontogenesis (M±m, %, n=10)

Age	Theophylline-resistant T-cells / Theophylline-sensitive T-cells		
	LDMR	**MDMR**	**HDMR**
0 hours	4,50±0,79	1,00±0,25	0
	9,30±0,34	0,10±0,11	0
4 hours	9,40±0,78**	2,70±0,45*	1,00±0,18***
	10,30±0,89	2,20±0,29***	0,40±0,11**
3 days	13,20±0,84***	2,60±0,54*	0,70±0,14***
	10,00±0,53	2,20±0,38***	0,50±0,18*
7 days	16,00±1,31***	3,50±0,53**	1,00±0,18***
	10,80±0,72	3,40±0,45***	0,70±0,14***
14 days	13,30±0,68***	4,20±0,45***	0,80±0,14***
	9,00±0,77	3,20±0,34***	0,30±0,18*
23 days	17,50±1,09***	5,20±0,76***	1,40±0,21***
	12,10±0,89*	4,10±0,37***	1,00±0,25**

Note: * – p≤0.05; ** – p≤0.01; *** – p≤0.001 compared to piglets at 0 hours of age.

So, for cells with low and medium density of membrane receptors in the theophylline-resistant lymphocytes, the correlation coefficient (r) was 0.99, while for cells with low density it was 0.98 compared to the homologous fractions of total T-lymphocytes. A high correlation coefficient value was found between changes in the content of T-suppressors and total T-lymphocytes with medium and high density of membrane receptors. The correlation coefficient (r) was 0.99 and 0.97, respectively. It should be noted that the number of theophylline-sensitive cells with low density of membrane receptors remained unchanged during the first weeks of life. Therefore, quantitative changes in the fraction of theophylline-resistant T-lymphocytes occurred due to cells with low, medium, and high density of membrane receptors, while changes in the fraction of theophylline-sensitive cells occurred only through an increase in the fraction of lymphocytes with medium and low density of membrane receptors.

Consumption of the «EHCW»preparation by sows contributed to an increase in lymphocytes in newborn piglets and provided significantly higher content of their T-, B-, and NK-forms during the first week of life (Table 10).

Table 10

Lymphocytes count in the blood of early postnatal piglets and its correction with the «EHCW»preparation (M±m, G/L, n = 10)

Age		Lymphocytes			
		T-	B-	NK-	0-
0 hours	control	0,20±0,02	0,13±0,02	0,06±0,02	0,96±0,13
	«EHCW»	0,36±0,06*	0,25±0,05*	0,14±0,02*	1,51±0,18*
4 hours	control	0,49±0,10	0,35±0,09	0,14±0,02	0,89±0,14
	«EHCW»	1,31±0,07***	0,83±0,10**	0,40±0,05**	1,38±0,16*
3 days	control	1,01±0,19	0,69±0,11	0,25±0,06	1,52±0,27
	«EHCW»	1,69±0,25*	1,05±0,10*	0,47±0,07*	1,35±0,18
7 days	control	1,56±0,21	0,93±0,09	0,43±0,07	1,52±0,23
	«EHCW»	3,29±0,30**	1,98±0,14***	0,93±0,08**	1,84±0,31
14 days	control	1,65±0,19	1,06±0,15	0,48±0,08	2,21±0,29
	«EHCW»	1,90±0,20	1,19±0,13	0,54±0,02	1,31±0,12*
23 days	control	2,53±0,20	1,38±0,16	0,54±0,08	1,70±0,18
	«EHCW»	3,03±0,15	1,79±0,09*	0,69±0,07	1,47±0,11

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to control.

It was found that prior to colostrum consumption, piglets from the experimental group of sows had a higher number of T-, B-, NK-, and 0-lymphocytes, respectively, by 1.80, 1.92, 2.33, and 1.57 times ($p \leq 0.05$) compared to the indicators in animals from the control group. The obtained results showed that the increase in the fraction of T-lymphocytes primarily occurred due to an increase in the number of cells with low and medium density of membrane receptors by 1.74 ($p \leq 0.05$) and 3.0 times, respectively (Table 11).

Table 11

Number of T-lymphocytes with different receptor density in the blood of piglets during the early postnatal ontogenesis and its correction with the «EHCW» preparation (M±m, G/L, n = 10)

Age		T-lymphocytes		
		LDMR	MDMR	HDMR
0 hours	control	0,19±0,02	0,01±0,01	0
	«EHCW»	0,33±0,06*	0,03±0,01	0
4 hours	control	0,37±0,07	0,09±0,02	0,03±0,01
	«EHCW»	0,98±0,05***	0,26±0,02**	0,06±0,01**
3 days	control	0,80±0,14	0,17±0,04	0,04±0,02
	«EHCW»	1,30±0,16*	0,32±0,06	0,08±0,02
7 days	control	1,18±0,15	0,30±0,06	0,07±0,01
	«EHCW»	2,44±0,19***	0,69±0,10*	0,16±0,03*
14 days	control	1,20±0,15	0,39±0,04	0,06±0,01
	«EHCW»	1,36±0,17	0,48±0,04	0,07±0,01
23 days	control	1,81±0,15	0,57±0,08	0,14±0,02
	«EHCW»	2,15±0,15	0,70±0,04	0,18±0,02

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to control.

After 4 hours of colostrum suckling, the difference between the control and experimental animals in the content of differentiated lymphocyte forms significantly increased. Thus, the fractions of T-, B-, and NK-lymphocytes in piglets from the experimental group were higher than those in the control group by 2.67 ($p \leq 0.001$), 2.37, and 2.86 ($p \leq 0.01$) times, respectively. The difference between the groups in the level of 0-lymphocytes remained almost unchanged and was 55.06% ($p \leq 0.05$) compared to the indicators in animals from the control group.

The number of B-lymphocytes in piglets from the experimental group increased only due to cells with low density of membrane receptors, the fraction of which was higher by 92.31% compared to the control group (Table 12).

Table 12

Number of B-lymphocytes with different receptor density in pig blood during the early postnatal ontogenesis period and its correction with the «EHCW» preparation (M±m, G/L, n = 10)

Age		**B-lymphocytes**		
		LDMR	**MDMR**	**HDMR**
0 hours	control	0,13±0,02	0,01±0,001	0
	«EHCW»	0,25±0,05*	0,01±0,001	0
4 hours	control	0,30±0,08	0,03±0,01	0,01±0,01
	«EHCW»	0,72±0,08**	0,09±0,02*	0,02±0,01
3 days	control	0,61±0,10	0,06±0,01	0,02±0,01
	«EHCW»	0,92±0,10*	0,10±0,01*	0,03±0,01
7 days	control	0,81±0,08	0,08±0,01	0,03±0,01
	«EHCW»	1,69±0,12***	0,20±0,02**	0,09±0,02*
14 days	control	0,95±0,12	0,09±0,02	0,02±0,01
	«EHCW»	1,04±0,11	0,10±0,02	0,05±0,02
23 days	control	1,18±0,12	0,14±0,03	0,06±0,02
	«EHCW»	1,48±0,09	0,22±0,02	0,09±0,02

Note: * – p≤0.05; ** – p≤0.01; *** – p≤0.001 compared to the control.

The changes in the number of T- and B-lymphocytes occurred simultaneously due to cells with different membrane receptor density. Thus, in the lymphocyte fraction of the experimental group, the number of T- and B-cells with low receptor density likely exceeded the values of control animals by 2.65 (p≤0.001) and 2.40 (p≤0.01) times, respectively, and in terms of the number of intermediate-avidity cells, it exceeded by 2.89 (p≤0.01) and 3.00 (p≤0.05) times, while for high-avidity lymphocytes, it was 2.0 times higher.

The difference between piglets in terms of T-, B-, and NK-lymphocytes decreased on average by almost 2.5 times on the 3rd day of life. The population of T-lymphocytes in the blood of the experimental pigs of this age exceeded the controls by 67.33% (p≤0.05), while the populations of B- and NK-lymphocytes exceeded by 52.17% and 88.00% (p≤0.05), respectively. The level of 0-form lymphocytes in the blood of the experimental animals remained almost unchanged compared to the previous value, while in the blood of the control group piglets, it increased by more than 70%, which was 11.18% higher than the value in the experimental group pigs.

The increase in the number of T-lymphocytes in 3-day-old piglets of the experimental group occurred against the background of a likely increase in the number of low-avidity cells by 62.50% ($p \leq 0.05$), intermediate-avidity cells by 88.24%, and high-avidity cells by 2.0 times compared to the values in the animals of the control group. At the same time, the fraction of B-lymphocytes increased mainly due to an increase in the number of low- and intermediate-avidity lymphocytes by 50.82% and 66.67% ($p \leq 0.05$), respectively, compared to the values in the animals of the control group. By the end of the first week, the level of each lymphocyte fraction in the blood of the experimental piglets exceeded the values in the control pigs. Thus, the difference in T-lymphocyte content was 2.11 times ($p \leq 0.01$), for B-lymphocytes – 2.13 times ($p \leq 0.001$), for NK-lymphocytes – 2.16 times, and for 0-lymphocytes – 1.21 times.

It was found that under the corrective action of the experimental preparation in 7-day-old piglets of the experimental group, the fractions of low-, intermediate-, and high-avidity T-lymphocytes were likely higher by 2.07 ($p \leq 0.001$), 2.30, and 2.29 times, respectively, and B-lymphocytes were higher by 2.09 ($p \leq 0.001$), 2.50 ($p \leq 0.01$), and 3.00 ($p \leq 0.05$) times compared to the values in the animals of the control group. In piglets at 14 and 23 days of life, there was a tendency to increase the fraction of T- and NK-lymphocytes, which was reflected in the lymphocyte predominance of these classes in the blood of the experimental animals by 15.15% and 12.50%, and 19.76% and 27.78%, respectively, compared to the values in the animals of the control group. Additionally, in these pigs, the fraction of B-lymphocytes was likely higher by 29.71% ($p \leq 0.05$) compared to the control group of animals.

It should be noted that during the first week of life, the fractions of low- and intermediate-avidity cells in the T-lymphocyte population accounted for an average of 75% and 20%, respectively, while in the B-lymphocyte population, they accounted for 85% and 10%, respectively. Therefore, the changes observed in these populations mainly occurred due to cells with low and intermediate membrane receptor density.

These changes were caused by the activation and maturation of the piglets' immune system after birth, thanks to the immunobiological and hormonal substances in colostrum and the rapid antigenic load[56, 57]. This indicates that not only colostral antibodies but also leukocytes and

---

[56]Inoue R., Tsukahara T. Composition and physiological functions of the porcine colostrum. *Animal science journal = Nihon chikusan Gakkaiho.* 2021. Vol. 92, No. 1. P. e13618.

[57] Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677.

biologically active substances that trantbocate with the mother's colostrum are involved in the formation of nonspecific immunity in newborn piglets[58]. Immunocorrection of the sows in the second half of gestation with the «EHCW» preparation contributed to an increase in the average number of T- and B-lymphocytes in the blood of newborn piglets during the first 23 days of life by 75% due to cells with low and intermediate membrane receptor density, and NK-lymphocytes by over 90%. These results suggest an indirectly stimulating effect of the experimental «EHCW»" preparation on the formation of cellular immune defense mechanisms in piglets and the enhancement of their resistance level.

According to the obtained results, it was found that «EHCW» promotes an increase in the fraction of T-lymphocytes, both resistant and sensitive to theophylline (Table 13 and 14).

Table 13

Number of T-helpers with different density levels of membrane receptors in the blood of piglets during early postnatal ontogenesis and its correction with «EHCW» preparation (M±m, G/L, n = 10)

Age		T-helpers			
		Total	**LDMR**	**MDMR**	**HDMR**
0 hours	control	0,07±0,01	0,06±0,01	0,01±0,01	0
	«EHCW»	0,15±0,03*	0,13±0,02*	0,02±0,01	0
4 hours	control	0,25±0,05	0,18±0,04	0,05±0,01	0,02±0,01
	«EHCW»	0,77±0,05***	0,56±0,04***	0,16±0,03**	0,05±0,01**
3 days	control	0,57±0,12	0,46±0,09	0,09±0,02	0,02±0,01
	«EHCW»	0,94±0,13*	0,70±0,08	0,20±0,04*	0,04±0,01
7 days	control	0,90±0,12	0,70±0,08	0,16±0,04	0,04±0,01
	«EHCW»	1,97±0,25**	1,46±0,15**	0,41±0,08*	0,11±0,03*
14 days	control	0,98±0,11	0,71±0,08	0,22±0,02	0,04±0,01
	«EHCW»	1,01±0,10	0,71±0,07	0,26±0,03	0,04±0,01
23 days	control	1,47±0,12	1,06±0,06	0,32±0,06	0,09±0,01
	«EHCW»	1,90±0,11*	1,40±0,09*	0,39±0,04	0,11±0,02

Note: * – p≤0.05; ** – p≤0.01; *** – p≤0.001 compared to the control.

[58]Chen Y., Tibbs-Cortes L. E., Ashley C., Putz A. M., Lim K. S., Dyck M. K., Fortin F., Plastow G. S., Dekkers J. C. M., Harding J. C. S., PigGen Canada. The genetic basis of natural antibody titers of young healthy pigs and relationships with disease resilience. *BMC genomics.* 2020. Vol. 21, No. 1. P. 648.

Indeed, before the intake of colostrum, the number of T-helper cells in the blood of piglets in the experimental group exceeded that of control animals by 2.14 times ($p \leq 0.05$), mainly due to a likely increase of cells with low membrane receptor density by 2.17 times ($p \leq 0.05$). After the colostrum consumption, this difference increased to 3.08 times ($p \leq 0.001$), with a simultaneous increase in the fractions of low, medium, and high-avid cells by 3.11 ($p \leq 0.001$), 3.20 ($p \leq 0.01$), and 2.50 ($p \leq 0.01$) times, respectively. However, at the age of 3 days, the difference decreased to 1.65 times.

Table 14

Number of T-suppressors with different density levels of membrane receptors in the blood of piglets during early postnatal ontogenesis and its correction with «EHCW» preparation (M±m, G/L, n = 10)

Age		T-suppressors			
		Total	**LDMR**	**MDMR**	**HDMR**
0 hours	control	0,13±0,02	0,12±0,02	0,01±0,001	0
	«EHCW»	0,21±0,04	0,20±0,04	0,01±0,001	
4 hours	control	0,24±0,05	0,19±0,03	0,04±0,01	0,01±0,001
	«EHCW»	0,54±0,04***	0,42±0,04**	0,10±0,01**	0,02±0,01
3 days	control	0,44±0,08	0,34±0,05	0,08±0,02	0,02±0,01
	«EHCW»	0,75±0,12*	0,59±0,09*	0,12±0,03	0,04±0,01
7 days	control	0,66±0,10	0,48±0,08	0,15±0,02	0,03±0,01
	«EHCW»	1,32±0,07***	0,99±0,05***	0,28±0,03*	0,05±0,01
14 days	control	0,67±0,09	0,49±0,08	0,17±0,02	0,01±0,01
	«EHCW»	0,90±0,11	0,65±0,10	0,22±0,02	0,03±0,01
23 days	control	1,06±0,11	0,75±0,10	0,25±0,02	0,06±0,01
	«EHCW»	1,13±0,06	0,75±0,06	0,30±0,02	0,08±0,03

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to the control.

It has been shown that in three-day-old piglets of the experimental group, the number of low-avid theophylline-resistant T-lymphocytes exceeded that of control animals by only 52.17%, while the number of medium-avid cells was likely to be 2.22 times higher ($p \leq 0.05$), and high-avid cells were 2.0 times higher.

By the end of the first week of life, the level of T-helper cells in the blood of the experimental pigs was 2.19 times higher ($p \leq 0.01$) compared to control animals, against a greater number of low, medium, and high-avid T-helper lymphocytes by 2.09 ($p \leq 0.01$), 2.56 ($p \leq 0.05$), and 2.75 ($p \leq 0.05$) times, respectively, based on the density of receptors on their plasma membrane.

No statistically significant difference was observed between the groups of 14-day-old piglets in terms of the content of theophylline-resistant lymphocytes. However, in piglets of the experimental group at 23 days of age, this subpopulation likely predominated over the values of the control piglets by 29.25% ($p \leq 0.05$) due to an increase in the number of low-avid forms by 32.08% ($p \leq 0.05$).

The differences between the experimental and control groups of animals in the number of theophylline-sensitive T-lymphocytes were similar to the changes observed in the theophylline-resistant cells. There was detected a tendency in respect with an increase the total number of T-suppressor cells in the blood of the experimental animals by 61.54% after the consumption of colostrum, which was accompanied by a 66.67% increase in low-avid cells.

A few hours after birth, the total number of theophylline-sensitive lymphocytes in the experimental group of pigs likely exceeded the values of the control pigs by 2.25 times ($p \leq 0.001$), against the backdrop of an increase in the fraction of low- and medium-avidity lymphocytes by 2.21 and 2.50 times, respectively ($p \leq 0.01$). This difference persisted until the end of the first week of life and was characterized by a 70.46% increase in the total T-suppressor cell count in the blood of the experimental pigs at 3 days of age, mainly due to low- and medium-avidity cells by 73.53% ($p \leq 0.05$) and 50.00%, respectively, and a 2.00-fold increase ($p \leq 0.001$) in 7-day-old animals, accompanied by an increase in the number of cells with low and medium membrane receptor density by 2.06 ($p \leq 0.001$) and 1.87 times ($p \leq 0.05$), compared to the indicators in the animals of the control group.

In 14-day-old animals of the experimental group, there was a tendency towards an increase in the fraction of theophylline-sensitive lymphocytes, manifested by a 34.33% higher level of these cells compared to the pigs in the control group, and by day 23 of life, the content of these lymphocytes did not differ significantly between the groups.

Research on the state of the immunoregulatory index in neonatal piglets (Figure 4) indicated that immediately after birth, this indicator had a low level (0.59 ± 0.11 AU). During the first week of life, the activation of the immune system in newborn piglets occurred, as evidenced by an increase in lymphocyte immunoregulatory index (LII) at 4 hours after suckling colostrum, as well as at 3 and 7 days of life, respectively, by 1.73, 2.20, and 2.36 times compared to the value of this indicator in newly born piglets. In the following sixty-five days of life, LII fluctuated within the range of 1.30-1.41 AU, which did not significantly differ from the value of LII in seven-day-old piglets.

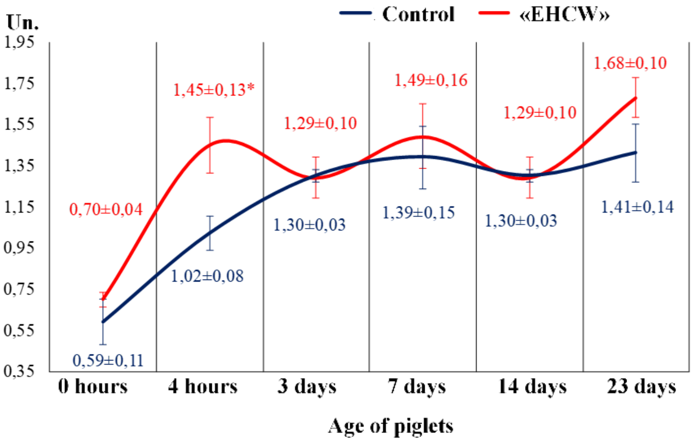

Fig. 4. LII in piglets during the early postnatal ontogenesis period and its correction with the «EHCW» preparation (M±m, n=10).

Note: * – p≤0.05 compared to control.

The immunocorrection of sow piglets with the «EHCW»preparation in the experimental group resulted in an increase in the LII indicator even before suckling colostrum by 18.64%. However, by the fourth hour of life, the difference between the groups increased to 42.15% with a probability of p≤0.05. Starting from the third day of life, the difference between the piglet groups in terms of the LII indicator was not significant.

Therefore, the immunocorrection of sow piglets with the «EHCW»preparation contributes to an increase in the fractions of theophylline-resistant and theophylline-sensitive lymphocytes in their piglets during the first 7 days of life, on average by 2.14 and 1.92 times, mainly due to an increase in low- and high-affinity cells. It also resulted in a 29.25% higher level of T-helper cells by the age of twenty-three days, against the background of changes in the fraction of cells with low receptor density, and led to a 42.15% increase in the LII level.

Thus, the early postnatal period of piglet ontogenesis was characterized by a high level of phagocytic activity of leukocytes in the blood of newborns in the first day of life, including activated oxygen-dependent mechanisms of bactericidal activity, and a gradual decrease in these cells in the second half of the first week of life by 7.7% and 16.0% (p≤0.05) respectively, and at the beginning of the fourth week by 11.3% and 29.8% (p≤0.01) respectively, against the background of an increase in the phagocytic number by 46.2% (p≤0.01) throughout the pre-weaning period and the phagocytosis completion index by 46.6% ($p \leq 0.05$) in the first 14 days compared to newborn animals, and a decrease in the LII by 17.7% from the fourteenth to the twenty-third day of life.

The state of neonatal piglets is accompanied by immunosuppression, as evidenced by the low value of the LII indicator (0.59 ± 0.11 AU), which is determined by the predominance of theophylline-sensitive T-lymphocytes in the blood of newly born piglets, along with a high level of 0-lymphocyte population. The 0-lymphocyte population is more representative in terms of the percentage of differentiated cells by 2.4 times and immune activation mechanisms in piglets from the first hours of life, against the background of an increase in the fraction of theophylline-resistant T-lymphocytes by 2.4 times in this period ($p \leq 0.01$), which contributes to an increase in the LII indicator during the first week of life.

The obtained results indicate the immaturity of the immune system in newborn pigs before consuming colostrum. This fact is determined by the influence of placental steroid hormones on the fetus, which exhibit immunosuppressive properties in the "mother-fetus" system. The hormonal influence on the fetal immune system promotes the enhanced formation of T-suppressors, along with the suppression of differentiation of T-helper cells and the antibody-forming capacity of B-lymphocytes[59, 60, 61].

Over time, during the early postnatal period, there is a redistribution of differentiated and 0-lymphocytes, accompanied by a gradual increase in the number of T and B-lymphocytes by 2.8 and 2.2 times respectively ($p \leq 0.001$) on the twenty-third day of life compared to animals before consuming colostrum, with a slight decrease by the end of the second week of life by 13.0% and 8.5% respectively compared to piglets at 7 days of life.

During the first week of the colostral period, the level of NK lymphocytes increased by 2.14 times ($p \leq 0.01$) compared to piglets without colostrum.

The administration of the «EHCW»preparation to the sows resulted in an increase in the following indicators in their piglets during the first 23 days of life: ALP by 11.5% ($p \leq 0.05$ – $p \leq 0.01$), NST level in the first week by 16.7% ($p \leq 0.05$), as well as an increase in FC by 22.4% ($p \leq 0.05$) on the seventh day

[59]Martinez C. A., Rubér M., Rodriguez-Martinez H., Alvarez-Rodriguez M. Pig Pregnancies after Transfer of Allogeneic Embryos Show a Dysregulated Endometrial/Placental Cytokine Balance: A Novel Clue for Embryo Death? *Biomolecules*. 2020. Vol. 10, No. 4. P. 554.

[60]Merlot E., Pastorelli H., Prunier A., Père M. C., Louveau I., Lefaucheur L., Perruchot M. H., Meunier-Salaün M. C., Gardan-Salmon D., Gondret F., Quesnel H. Sow environment during gestation: part I. Influence on maternal physiology and lacteal secretions in relation with neonatal survival. *Animal*. 2019. Vol. 13, Suppl 7. P. 1432–1439.

[61]Hoffmann J. P., Liu J. A., Seddu K., Klein S. L. Sex hormone signaling and regulation of immune function. *Immunity*. 2023. Vol. 56, No. 11. P. 2472–2491.

of life and PHA by 43.8% on the twenty-third day compared to the control group animals.

In the blood of piglets obtained from sows that were exposed to the «EHCW» preparation during the first week of life, a higher number of T-lymphocytes was observed, on average, 2.1 times higher due to an increase in theophylline-resistant and theophylline-sensitive lymphocytes, primarily with low and medium membrane receptor density, on average, 2.14 and 1.92 times ($p \leq 0.05$ – $p \leq 0.001$), and an increase in the number of B- and NK-lymphocytes, on average, 2.0 and 2.3 times ($p \leq 0.05$ – $p \leq 0.001$), against the background of an increase in the number of 0-lymphocytes by 1.56 times ($p \leq 0.05$) and LII by 42.15% ($p \leq 0.05$) on the first day.

3. Research on the formation of the humoral branch of immunity in the blood of piglets

The level of total protein and the ratio between protein fractions in the blood of piglets in the first days of life are closely related to the protein composition of the sow's colostrum[62]. The level of total protein in the blood of newborn piglets before colostrum intake was the lowest, measuring 25.46±0.82 g/L, and within four hours, it increased by 61.90% ($p \leq 0.01$), as presented in Figure 5.

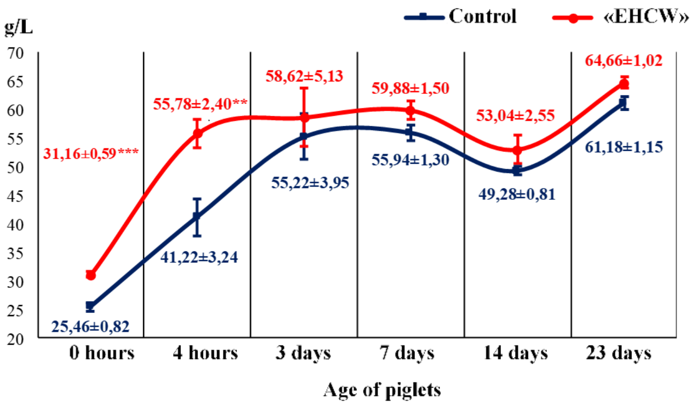

Fig. 5. Level of total protein in the blood of piglets during the early postnatal ontogenesis period and its correction with the «EHCW» preparation (M±m, n=10)

Note: 1. ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to the control.

[62]Inoue R., Tsukahara T. Composition and physiological functions of the porcine colostrum. *Animal science journal = Nihon chikusan Gakkaiho*. 2021. Vol. 92, No. 1. P. e13618.

On the 3rd and 7th day of life, the content of total protein was almost the same. At the same time, the level of total protein exceeded the values of one-day-old piglets by almost 34%, and compared to non-colostrum-fed piglets, it was significantly higher by 2.18 times ($p \leq 0.001$). In two-week-old piglets, there was a slight decrease in the level of total serum protein by 13.52% compared to one-week-old piglets, but the protein content in fourteen-day-old piglets was still significantly higher than non-colostrum-fed piglets by 93.56% ($p \leq 0.001$). Over the next 9 days of life, there was an increase in the level of total protein to 61.18±1.15 g/L, which was 2.40 times higher than the value in piglets before colostrum intake ($p \leq 0.001$).

According to the research results on protein fractions, it was demonstrated that before colostrum intake, the globulin fraction predominated in the blood of piglets, with a total quantity accounting for over 56% of the total protein. Within this group, approximately 53.46% was attributed to β-globulins, almost 35% in an equal proportion to α-2 and γ-fractions, and 11% to α-1 globulins (Table 15).

Table 15

Dynamics of protein fractions in the blood of piglets in the control group during the early postnatal ontogenesis period (M±m, %, n=10)

Age	Albumins	Globulins			
		α-1	α-2	β-	γ-
0 hours	43,45±1,54	6,22±0,88	9,74±0,60	30,23±0,89	10,36±0,89
4 hours	51,21±1,63**	10,47±1,15*	7,70±0,76*	13,03±1,41***	17,59±1,44**
3 days	44,42±5,60	7,64±2,68	4,76±1,29**	21,53±5,40*	21,64±0,60***
7 days	51,11±1,34**	5,14±0,96	7,67±0,87	16,40±2,10***	19,68±1,89**
14 days	53,30±1,97**	5,04±0,83	9,15±0,37	16,10±1,51***	16,41±1,36**
23 days	40,04±1,95	6,05±0,40	11,01±1,24	17,20±2,14***	25,70±1,40***

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$; *** – $p \leq 0.001$ compared to 0-hour-old piglets.

Four hours after colostrum suckling, a redistribution of protein fractions occurred in the blood of newborn piglets, resulting in a significant increase in albumin, α-1, and γ-globulin fractions by 17.86% ($p \leq 0.01$), 68.33%, and 69.79% ($p \leq 0.05$), respectively, while the α-2 and β-globulin fractions decreased by 20.95% ($p \leq 0.05$) and 56.90% ($p \leq 0.001$), respectively, compared to the values in animals at birth.

On the third day of life, a decrease was observed, almost returning to the initial level of albumin and α-1 globulin levels, along with a progressive decrease in the α-1 fraction by more than 50% ($p \leq 0.01$) compared to piglets before colostrum intake. These changes were accompanied by an increase in the percentage of β- and γ-globulin fractions by 65.23% and 23.02%, respectively, compared to the previous values. It should be noted that the γ-globulin fraction increased significantly by 2.09 times ($p \leq 0.001$), while the β-fraction remained nearly 30% lower than their values in animals at birth.

At the end of the first week and throughout the following 7 days, an increase in the albumin fraction by 17.63% and 22.67% ($p \leq 0.01$), respectively, and the α-2 globulin fraction by 61.13% and 92.23% was observed in the blood of piglets in the control group compared to three-day-old piglets. The γ-globulin fraction showed a reverse dynamic characterized by a decrease in the relative content of these proteins by 9.06% and 24.17%, respectively, compared to piglets on the third day of life. However, the amount of γ-globulins remained significantly higher compared to the values in animals at birth by 89.96% and 58.40% ($p \leq 0.01$), respectively.

On the seventh day of life, the relative amount of α-1 and β-globulin fractions decreased by 32.72% and 23.83%, respectively, compared to three-day-old piglets, and remained at this level until the end of the second week of life.

In piglets at twenty-three days of age, the highest level of the γ-globulin fraction was observed, which exceeded that of precolostrum piglets by almost 2.5 times ($p \leq 0.001$), and the lowest relative amount of albumins throughout the study. This was accompanied by an increase in the globulin fraction in the last 9 days to almost 60% due to an increase in γ- and α-2-globulins by 56.61% and 20.33%, respectively, along with a decrease in the albumin fraction by 24.9% compared to two-week-old piglets.

Thus, the early postnatal period in piglets was characterized by a low level of total protein immediately after birth, with globulins, mainly β-fractions, representing the majority. Subsequently, the total serum protein increased during the first three days of life, mainly due to the γ-globulin fraction, while the relative amount of α- and β-globulins decreased. Starting from the seventh to the fourteenth day of life, the amount of γ-globulins decreased, accompanied by an increase in the albumin fraction. At the beginning of the fourth week, the relative amount of globulins increased mainly due to the γ-globulin fraction, while albumins decreased.

The administration of the «EHCW»drug to the blood of newborn piglets in the experimental group, before and after colostrum intake, resulted in an increase in the level of total protein by 22.39% ($p \leq 0.001$) and 35.32% ($p \leq 0.01$), respectively. This increase was due to an elevation in the fractions of β-globulins by 27.50% ($p \leq 0.01$) and 56.96% ($p \leq 0.05$), and γ-globulins by 80.68% ($p \leq 0.01$) and 50.07% ($p \leq 0.05$), compared to the indicators in the animals of the control group (Table 16).

Table 16

Protein fractions of piglets during the early postnatal ontogenesis period and their correction with the «EHCW»drug (M±m, g/L, n = 10)

Age		Albumins	Globulins			
			α-1	**α-2**	**α-1**	**γ-**
0 hours	control	11,04±0,38	1,59±0,26	2,47±0,13	7,71±0,40	2,64±0,26
	«EHCW»	12,14±0,46	2,00±0,18	2,42±0,24	9,83±0,27**	4,77±0,46**
4 hours	control	21,11±1,78	4,32±0,66	3,18±0,38	5,46±0,89	7,15±0,49
	«EHCW»	27,43±1,25*	4,67±0,14	4,39±0,66	8,57±0,42*	10,73±1,12*
3 days	control	24,24±2,80	4,49±1,82	2,67±0,74	11,86±3,05	11,95±0,91
	«EHCW»	23,87±3,04	3,25±0,58	2,46±0,49	11,40±1,83	17,65±2,13*
7 days	control	28,58±0,88	2,86±0,52	4,29±0,52	9,21±1,29	11,00±1,06
	«EHCW»	27,31±3,00	2,16±0,46	4,57±0,78	7,15±0,80	18,69±2,46*
14 days	control	26,26±0,98	2,50±0,44	4,51±0,24	7,91±0,69	8,10±0,73
	«EHCW»	27,35±0,69	2,33±0,52	5,24±0,50	7,19±0,69	10,93±0,90*
23 days	control	24,51±1,38	3,70±0,28	6,71±0,68	10,58±1,42	15,68±0,62
	«EHCW»	28,65±0,79*	3,29±0,36	5,63±0,65	9,49±0,80	17,59±0,69

Note: * – $p \leq 0.05$; ** – $p \leq 0.01$ compared to control.

It was demonstrated that after colostrum intake in piglets of the first day of life, the level of albumin likely increased by almost 30% ($p \leq 0.05$) compared to the indicators in the animals of the control group.

No significant difference was found between the control and experimental groups of piglets regarding the level of total protein. However, the quantity of total protein in the experimental group animals was on average 6.6% higher than the values in the control piglets throughout the entire research period, due to a likely increase in the amount of γ-globulins in piglets at 3, 7, and 14 days of life by 47.70%, 69.91%, and 34.94% ($p \leq 0.05$), respectively.

On the 23rd day of life, piglets in the experimental group showed an increase in the albumin content by 16.89% ($p \leq 0.05$). During this time, there was also a tendency for an increase in the γ-globulin fraction by 12.18% compared to the control animals.

Thus, immunocorrection of sow piglets with the «EHCW»drug contributes to an increase in the total protein level in daily piglets through albumins, β- and γ-globulins.

The results of the study on the indicators of the humoral immune response indicate that the serum of newborn piglets had the lowest level of immune protection before colostrum intake. This was reflected in the low levels of BASB and LASB indicators, along with a slight amount of CIC (Table 17).

Table 17

Level of BASB, LASB, and CIC in piglet blood during the early postnatal ontogenesis period and their correction with the «EHCW»drug (M±m, n=10)

Age		BASB, %	LASB, %	CIC, Un.
0 hours	control	18,31±1,10	2,17±0,14	11,80±1,08
	«EHCW»	22,48±1,17*	2,19±0,24	11,40±0,76
4 hours	control	27,35±2,07^{00}	9,28±1,07^{000}	32,00±3,45^{000}
	«EHCW»	35,00±1,62*	13,52±1,38*	30,60±1,52
3 days	control	34,68±2,91^{000}	22,37±1,26^{000}	65,60±4,83^{000}
	«EHCW»	42,65±1,87*	26,65±0,71*	34,80±2,38***
7 days	control	36,86±1,68^{000}	22,52±1,26^{000}	51,40±4,58^{000}
	«EHCW»	42,89±1,91*	24,01±1,29	33,80±3,19*
14 days	control	28,13±1,41^{000}	23,47±1,50^{000}	42,40±5,16^{000}
	«EHCW»	33,40±1,60*	23,46±1,32	31,20±3,13
23 days	control	70,60±1,49^{000}	24,95±0,97^{000}	52,00±3,54^{000}
	«EHCW»	52,63±3,13	24,40±1,29	48,80±2,84

Note: 1. 00 – p≤0.01; 000 – p≤0.001 compared to 0-hour-old piglets; 2. * – p≤0.05; *** – p≤0.001 compared to control.

Modulation of the bactericidal activity of piglet serum during the early postnatal period was characterized by a gradual increase during the first week of life, a decrease in two-week-old animals, and a sharp increase in the fourth week after birth.

The BASB level increased by 49.37% (p≤0.01) in the first four hours of piglets' life compared to animals before suckling colostrum. On the third day, this indicator increased by 26.80% compared to the previous value and was significantly higher by 89.41% (p≤0.001) compared to precolostral piglets. Over the next four days, the BASB increased by only 6.29% compared to animals of three-day-old age, while the level of this indicator in seven-day-old piglets was more than twice as high ($p \leq 0.001$) as in piglets before colostrum intake.

Considering that the acquired immune system in newborn animals is more complex and requires some time for its implementation, in the first minutes of life, the organism of newborns is protected by components of the nonspecific resistance system. They receive these components from the mother's colostrum. The immune components of colostrum, upon entering the newborn's body, react to particulate agents and toxic substances that come into contact with the piglets' immune system. Therefore, suckling colostrum from the first hours of life increases the level of humoral defense mechanisms in piglet blood. This is evidenced by the increase in the γ-globulin fraction, the BASB indicator, lysozyme activity, and the number of CIC[63, 64, 65].

By the end of the second week of life, piglets showed a 23.68% decrease in BASB compared to seven-day-old animals, but compared to piglets without colostrum intake, this indicator remained significantly higher by 53.63% ($p \leq 0.001$). In the following 9 days, there was a sharp increase of 2.51 times in the BASB indicator compared to the value in two-week-old pigs and 3.86 times ($p \leq 0.001$) compared to piglets before colostrum intake. Since the bactericidal activity of serum is manifested due to the presence of humoral immune components such as immunoglobulins, the complement system, and lysozyme, a decrease in the BASB indicator may be due to a decrease in the absolute quantity of γ-globulins.

The lysozyme activity of piglet serum (LASB) in the early neonatal period was characterized by a low level in newborn pigs before colostrum intake and a rapid increase during the first three days of life. The LASB indicator in piglets after four hours of birth significantly increased by 4.28 times ($p \leq 0.001$), and on the third day of life, it was more than 10 times higher ($p \leq 0.001$) compared to precolostral animals, maintaining this level until the end of the first week of life.

In two-week-old piglets, the LASB indicator increased by almost 5% compared to animals at three days of age, and by the twenty-third day, this difference reached 11.37%.

---

[63] Orije M.R.P., Maertens K., Corbière V., Wanlapakorn N., Van Damme P., Leuridan E., Mascart F. The effect of maternal antibodies on the cellular immune response after infant vaccination: A review. *Vaccine*. 2020. Vol. 38, No. 1. P. 20–28.

[64] Chepngeno J., Amimo J. O., Michael H., Jung K., Raev S., Lee M. V., Damtie D., Mainga A. O., Vlasova A. N., Saif L. J. Rotavirus A Inoculation and Oral Vitamin A Supplementation of Vitamin A Deficient Pregnant Sows Enhances Maternal Adaptive Immunity and Passive Protection of Piglets against Virulent Rotavirus A. *Viruses*. 2022. Vol. 14, No. 11. P. 2354.

[65] Krogh U., Quesnel H., Le Floch N., Simongiovanni A., van Milgen J. A dynamic mammary gland model describing colostrum immunoglobulin transfer and milk production in lactating sows. *Journal of animal science*. 2021. Vol. 99, No. 2. P. skab030.

The research results regarding the content of circulating immune complexes (CIC) in the blood of neonatal piglets showed a relatively low quantity in precolostral animals and a significant increase of 2.71 times ($p \leq 0.001$) after four hours of birth and 5.56 times ($p \leq 0.001$) on the third day of life compared to precolostral pigs.

During the following eleven days, a downward trend in CIC quantity was observed, characterized by a decrease of 21.65% in seven-day-old pigs compared to three-day-old piglets and 17.56% in two-week-old animals compared to seven-day-old pigs. In this context, the level of CIC in the blood of seven and fourteen-day-old piglets remained significantly higher, 4.36 and 3.59 times respectively, compared to animals before colostrum intake.

The piglets showed a 22.64% increase in CIC (complement immune complex) levels in their blood on the twenty-third day of life compared to animals at two weeks of age. Thus, the neonatal period was characterized by low levels of BASB (bactericidal activity of serum complement), LASB (lysozyme activity of serum complement), and CIC prior to colostrum intake, followed by a sharp increase within the first day of life. The early postnatal period of pig ontogenesis was accompanied by an elevation of BASB and LASB levels during the first twenty-three days, with a slight decrease in BASB at two weeks of age, followed by a sharp increase at the beginning of the fourth week of life, and a significant increase in LASB during the first three days after birth and gradual elevation throughout the pre-weaning period.

The dynamics of CIC in neonatal animals were characterized by a rapid increase until the third day of life, followed by a decrease until the end of the second week after birth, after which a tendency for a subsequent increase in this indicator was observed.

Immunocorrection of the sow's immune system using the «EHCW»preparation resulted in an enhancement of humoral immune defense indicators in piglets during the first two weeks of life and a decrease in CIC levels during this period.

The analysis of BASB indicators in piglets revealed a 22.77% increase ($p \leq 0.05$) in bactericidal activity levels in piglets before colostrum intake. After four hours of birth, this difference increased to 27.97% ($p \leq 0.05$). On the third day of life, the BASB indicator was significantly higher in the experimental group of piglets by nearly 23% ($p \leq 0.05$) compared to the control animals.

The probable difference in the BASB indicator between the groups persisted during the first two weeks of life and was evident on the seventh and fourteenth days of life with values of BASB in the blood of experimental piglets higher by 16.36% and 18.73% ($p \leq 0.05$) respectively compared to the control animals.

Unlike bactericidal activity, the lysozyme activity of blood serum in piglets did not differ significantly between the groups before colostrum intake. However, after a few hours of life, the LASB indicator in the experimental piglets significantly exceeded the values of the control animals by 45.69% ($p \leq 0.05$). On the third day of life, this difference decreased to 19.13% but remained statistically significant ($p \leq 0.05$).

The level of CIC in the blood of piglets on the first day of life did not differ significantly between the experimental and control groups. A significant difference in this indicator was observed in pigs at the age of 3 and 7 days. At this time, the CIC level in the blood of experimental piglets was lower than that of the control animals by 46.95% ($p \leq 0.001$) and 34.24% ($p \leq 0.05$) respectively.

On the 14th day of life, no significant difference in CIC levels between the groups was detected, but there was a tendency for a decrease in this indicator in the blood of the experimental piglets by 26.42% compared to the control animals. The decrease in CIC levels in piglet blood is associated with the elimination of antigens due to the increased activity of macrophages containing Toll-like transmembrane receptors. These receptors play a key role in innate immunity by activating and regulating cellular immune responses through conjugation with antigenic determinants[66, 67].

At the end of the experiment, the difference between the groups ranged within 6% and was not statistically significant.

So, immunocorrection of sows using the «EHCW»preparation contributed to an increase in the average LASB index in their piglets by 32.41% in the first 3 days of life and an average BASB level increase by 22.52% during the first two weeks. Additionally, the number of CIC decreased by 46.95%, 34.24%, and 26.42% on days 3, 7, and 14 of life, respectively, compared to the indicators in the control group animals.

Based on the above, it can be concluded that the early postnatal period in piglets was characterized by a low level of total protein in pig blood before colostrum intake (25.46±0.82 g/L), of which globulins accounted for the majority (56.55%), mainly the β-fraction. This resulted in low BASB (18.31±1.10%), LASB (2.17±0.14%), and CIC (11.80±1.08 Ohm) levels during this period. Over the next three days of life, the total serum protein

[66] Aegerter H., Lambrecht B. N., Jakubzick C. V. Biology of lung macrophages in health and disease. *Immunity*. 2022. Vol. 55, No. 9. P. 1564–1580.

[67] Hăbeanu M., Lefter N. A., Gheorghe A., Ropota M., Toma S. M., Pistol G. C., Surdu I., Dumitru M. Alterations in Essential Fatty Acids, Immunoglobulins (IgA, IgG, and IgM), and Enteric Methane Emission in Primiparous Sows Fed Hemp Seed Oil and Their Offspring Response. *Veterinary sciences*. 2022. Vol. 9, No. 7. P. 352.

increased 2.2 times ($p \leq 0.001$), primarily due to the γ-globulin fraction, which increased 2.1 times ($p \leq 0.001$), while the relative quantity of α-2- and β-globulins decreased by 2.1 ($p \leq 0.01$) and 1.4 times ($p \leq 0.05$), respectively, compared to the indicators at birth.

Starting from the seventh to the fourteenth day of life in pigs, a decrease in the γ-globulin level by 9.1% and 24.2% was observed, accompanied by an increase in albumin levels by 15.1% and 20.0%, respectively, compared to piglets at 3 days old.

By the beginning of the fourth week, the globulin level increased by 28.4%, mainly due to a 56.6% increase in the γ-globulin fraction compared to animals at 14 days old. During this time, the γ-globulin level was 2.5 times higher ($p \leq 0.001$) compared to the indicators at birth.

The early postnatal period of pig ontogenesis was accompanied by an increase in BASB and LASB indicators during the first twenty-three days by 3.9 times ($p \leq 0.001$) and 11.2 times ($p \leq 0.001$), respectively, with a slight decrease in BASB at the two-week age.

The dynamics of CIC content in pig blood during the neonatal period were characterized by a 5.6-fold increase ($p \leq 0.001$) on the third day of life compared to precolostral piglets, followed by a decrease on the seventh, fourteenth, and twenty-third day by 21.6%, 35.4%, and 20.7%, respectively, compared to the values of piglets at 3 days old.

Immunocorrection of sows using the «EHCW» preparation contributed to an average increase of 28.9% ($p \leq 0.01$ – $p \leq 0.001$) in the total protein level in piglets within the first day of life, primarily due to an increase in the β- and γ-globulin fractions by 43.9% and 65.4% ($p \leq 0.05$ – $p \leq 0.01$), respectively. It also resulted in an increase of γ-globulins during the first two weeks of life by 50.8% ($p \leq 0.05$), while during this time, the BASB level increased by an average of 21.8% ($p \leq 0.05$) compared to the indicators in the control group animals.

The application of the «EHCW»preparation to sows led to an increase in the lysozyme activity in the blood serum of their piglets during the first 3 days after birth by an average of 32.4% ($p \leq 0.05$) and a decrease in CIC on days 3 and 7 of life by 47.0% ($p \leq 0.001$) and 34.2% ($p \leq 0.05$), respectively.

The use of «EHCW»led to an increase in the number of leukocytes, including lymphocytes, in the blood of lactating sows, which resulted in an increase in the γ-globulin fraction due to the synthesis of immunoglobulins, as well as BASB and LASB indicators. Additionally, enrichment of immune components in the colostrum of the experimental group sows was observed, which contributed to the enhancement of its immunobiological properties compared to the control animals[68]. The consumption of such colostrum by piglets resulted in an increase in the number of T-lymphocytes, primarily the theophylline-resistant and theophylline-sensitive lymphocytes, with low and moderate membrane receptor density, as well as the level of B- and NK-lymphocytes, along with an increase in the fraction of 0-lymphocytes within the first day[69].

CONCLUSIONS

It was proved that the early piglets postnatal ontogeny period is characterized by sufficiently high rates of red blood cells, hemoglobin and hematocrit levels at birth and their significant decrease during the first seven days of life. The application of «EHCW» increases ($p \leq 0.05$) the hemoglobin level by 16.6% and the number of erythrocytes by 9.9%, and normalizes hematocrit level in piglets during the first 7 days of life compared to animals of control.

It is noted that the absolute amount of leukocytes in the blood of newly born piglets is low (3,32 ± 0,28 G/dm3) and polymorphonuclear cells predominate. During the first 7 days of the piglets life the number of leukocytes in their blood increases due to lymphocytes. Application of «EHCW» increases the amount of leukocytes by 1.5 times in the blood of piglets during the first 7 days of life, but does not exceed the limits of physiological values. These changes are due to an increase number of lymphocytes and segmental neutrophils, respectively by 1.7 and 1.3 times compared to piglets of control.

The analysis of cellular and humoral portions of piglets immunity in the period of early postnatal ontogenesis indicates that newborns have low immunological parameters prior to the consumption of colostrum, (except phagocytic leukocytes) and gradually increase to 23 days of life. The use of «EHCW» for sows increases the number of 0-lymphocytes in the blood of piglets in the first day of life by 1.6 times. Subsequently, during the first week of postnatal development of piglets, the number of T-, B- and NK-lymphocytes an increases by 2.1, 2.1 and 2.3 times. The number of phagocytic leukocytes with an activated oxygen dependent bacteriolytic mechanism also increases by 16.7%, aggressiveness and digestibility

[68] Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677.

[69] Inoue R., Tsukahara T. Composition and physiological functions of the porcine colostrum. *Animal science journal = Nihon chikusan Gakkaiho*. 2021. Vol. 92, No. 1. P. e13618.

increased by 22.4% and 43.8%, level of phagocytic activity of leukocytes by 11.5% ($p \leq 0.05$) compared to the animals of the control.

The use of «EHCW» increases the level of γ-globulins in the first 14 days by 56.7% ($p \leq 0.05$ – $p \leq 0.01$), BASB by 21.8%, decreases the level of CIC by 40.6% during the first 7 days of life and the activity of lysozyme by 32.4% in the first 3 days after birth compared to the control.

SUMMARY

Blood parameters confirmed as markers to assess various physiological and biochemicals processes including neonatal development of farming animals. Consequently, the effects of natural compounds can reflect both protective and detrimental effect on the swine and piglet health. The stimulation of piglet viability during early neonatal period is remain actual item of swine farming. The maintenance of immune resistance including both humoral and cellular potential is one of prospective manner to reduce the mortality in piglet litters. Immunimodulatory formulations based on natural chemicals correspond to this purpose. Present study demonstrated the beneficial effect of enzyme hydrolyzed cell wall of Lactobacillus (EHCW) on haematological parameters of piglets including hemoglobin content (+16.6%) and erythrocyte shape. The exposure to EHCW induced statistical increase in lymphocytes and segmented neutrophils number. Additionally, «EHCW» application stimulates an increase in active phagocyte content and its digestive capability. The analysis of cellular and humoral immunity during early postnatal ontogenesis demonstrated that weak immune potential of control piglet group could be modulated with «EHCW» exposure. The application of «EHCW» increased the number of 0-lymphocytes in the blood of piglets in the first day of life by 1.6 times. Subsequently, during the first week of postnatal development of piglets, the number of T-, B- and NK-lymphocytes an increased by 2.1, 2.1 and 2.3 times accordingly. The number of phagocytic leukocytes with an activated oxygen dependent bacteriolytic mechanism also increases by 16.7%, aggressiveness and digestibility increased by 22.4% and 43.8%, level of phagocytic activity of leukocytes by 11.5% ($p \leq 0.05$) compared to the animals of the control. Obtained results evidence that fragments of Lactobacillus cell wall are the prospective tool to modulate the neonatal piglet immunity and maintain viability of piglet litters.

Bibliography

1. Abrao Trad A. T., Buddington R., Enninga E., Duncan J., Schenone C. V., Mari G., Buddington K., Schenone M. Report of an Experiment With a Fetal

Ex-Utero Support System in Piglets. *Cureus*. 2023. Vol. 15, No. 4. P. e38223. DOI: 10.7759/cureus.38223

2. Aegerter H., Lambrecht B. N., Jakubzick C. V. Biology of lung macrophages in health and disease. *Immunity.* 2022. Vol. 55, No. 9. P. 1564–1580. DOI: 10.1016/j.immuni.2022.08.010

3. Annacker O., Pimenta-Araujo R., Burlen-Defranoux O., Bandeira A. On the ontogeny and physiology of regulatory T cells. *Immunological reviews*. 2001. Vol. 182. P. 5–17. DOI: 10.1034/j.1600-065x.2001.1820101.x

4. Bidarimath M., Tayade C. Pregnancy and spontaneous fetal loss: A pig perspective. *Molecular reproduction and development*. 2017. Vol. 84, No. 9. P. 856–869. DOI: 10.1002/mrd.22847

5. Bonilla M.C., Fingerhut L., Alfonso-Castro A., Mergani A., Schwennen C., von Köckritz-Blickwede M., de Buhr N. How Long Does a Neutrophil Live?-The Effect of 24 h Whole Blood Storage on Neutrophil Functions in Pigs. *Biomedicines*. 2020. Vol. 8, No. 8. P. 278. DOI: 10.3390/biomedicines8080278

6. Branch D. W. Physiologic adaptations of pregnancy. *Am J Reprod Immunol*. 1992. Vol. 28, No. 3–4. P. 120–122. DOI: 10.1111/j.1600-0897.1992.tb00771.x

7. Bréa D., Soler L., Fleurot I., Melo S., Chevaleyre C., Berri M., Labas V., Teixeira-Gomes A. P., Pujo J., Cenac N., Bähr A., Klymiuk N., Guillon A., Si-Tahar M., Caballero I. Intrinsic alterations in peripheral neutrophils from cystic fibrosis newborn piglets. *Journal of cystic fibrosis: official journal of the European Cystic Fibrosis Society.* 2020. Vol. 19, No. 5. P. 830–836. DOI: 10.1016/j.jcf.2020.02.016

8. Chen Y., Tibbs-Cortes L. E., Ashley C., Putz A. M., Lim K. S., Dyck M. K., Fortin F., Plastow G. S., Dekkers J. C. M., Harding J. C. S., PigGen Canada. The genetic basis of natural antibody titers of young healthy pigs and relationships with disease resilience. *BMC genomics.* 2020. Vol. 21, No. 1. P. 648. DOI: 10.1186/s12864-020-06994-0

9. Cheng Y., Azad M. A. K., Ding S., Liu Y., Blachier F., Ye T., Kong X. Metabolomics Analysis Reveals the Potential Relationship Between Sow Colostrum and Neonatal Serum Metabolites in Different Pig Breeds. *Molecular nutrition & food research*. 2023. Vol. 67, No. 16. P. e2200677. DOI: 10.1002/mnfr.202200677

10. Cheng Z., Zhou S. T., Zhang X. H., Fu Q., Yang Y., Ji W. B., Liu H. G. Effects of early intermittent maternal separation on behavior, physiological, and growth performance in piglets. *Journal of animal science.* 2023. Vol. 101. P. skad122. DOI: 10.1093/jas/skad122

11. Chepngeno J., Amimo J. O., Michael H., Jung K., Raev S., Lee M. V., Damtie D., Mainga A. O., Vlasova A. N., Saif L. J. Rotavirus A Inoculation

and Oral Vitamin A Supplementation of Vitamin A Deficient Pregnant Sows Enhances Maternal Adaptive Immunity and Passive Protection of Piglets against Virulent Rotavirus A. *Viruses*. 2022. Vol. 14, No. 11. P. 2354. DOI: 10.3390/v14112354

12. De Carvalho R. H., Callegari M. A., Dias C. P., Kirwan S., da Costa M. C. R., da Silva C. A. Euglena gracilis β-Glucans (1,3): Enriching Colostrum of Sow for Enhanced Piglet Immunity. *Animals: an open access journal from MDPI*. 2023. Vol. 13, No. 22. P. 3490. DOI: 10.3390/ani13223490

13. Dong L., Zhong X., Zhang L., Kong L., Kong Y., Kou T., Wang T. Impaired intestinal mucosal immunity is associated with the imbalance of T lymphocyte sub-populations in intrauterine growth-restricted neonatal piglets. *Immunobiology.* 2015. Vol. 220, No. 6. P. 775–781. DOI: 10.1016/j.imbio.2014.12.017

14. Farmer C., Edwards S. A. Review: Improving the performance of neonatal piglets. *Animal: an international journal of animal bioscience.* 2022. Vol. 16, Suppl 2. P. 100350. DOI: 10.1016/j.animal.2021.100350

15. Forner R., Bombassaro G., Bellaver F. V., Maciag S., Fonseca F. N., Gava D., Lopes L., Marques M. G., Bastos A. P. Distribution difference of colostrum-derived B and T cells subsets in gilts and sows. *PloS one.* 2021. Vol. 16, No. 5. P. e0249366. DOI: 10.1371/journal.pone.0249366

16. Grün V., Schmucker S., Schalk C., Flauger B., Weiler U., Stefanski V. Influence of Different Housing Systems on Distribution, Function and Mitogen-Response of Leukocytes in Pregnant Sows. *Animals: an open access journal from MDPI.* 2013. Vol. 3, No. 4. P. 1123–1141. DOI: 10.3390/ani3041123

17. Hăbeanu M., Lefter N. A., Gheorghe A., Ropota M., Toma S. M., Pistol G. C., Surdu I., Dumitru M. Alterations in Essential Fatty Acids, Immunoglobulins (IgA, IgG, and IgM), and Enteric Methane Emission in Primiparous Sows Fed Hemp Seed Oil and Their Offspring Response. *Veterinary sciences*. 2022. Vol. 9, No. 7. P. 352. DOI: 10.3390/vetsci9070352

18. Hedges J. F., Snyder D. T., Robison A., Thompson M. A., Aspelin K., Plewa J., Baldridge J., Jutila M. A. A TLR4 agonist liposome formulation effectively stimulates innate immunity and enhances protection from bacterial infection. *Innate immunity*. 2023. Vol. 29, No. 3-4. P. 45–57. DOI: 10.1177/17534259231168725

19. Hoffmann J. P., Liu J. A., Seddu K., Klein S. L. Sex hormone signaling and regulation of immune function. *Immunity.* 2023. Vol. 56, No. 11. P. 2472–2491. DOI: 10.1016/j.immuni.2023.10.008

20. Holda A. A., Yefimov V. H. Biochemical serum characteristics in fattening pigs infected with porcine circovirus type 2. *Theoretical and Applied Veterinary Medicine.* 2023. Vol. 11, No. 1. P. 15–21. doi: 10.32819/2023.11003

21. Holda K. O., Masiuk D. M., Kokariev A. V., Vasilenko T. O. Colostral immunity of piglets to the virus of Aujeszky's disease with active sows' immunization. *Theoretical and Applied Veterinary Medicine*. 2020. Vol. 8, No. 4. P. 257–260. https://doi.org/10.32819/2020.84037

22. Huang G., Li X., Lu D., Liu S., Suo X., Li Q., Li N. Lysozyme improves gut performance and protects against enterotoxigenic Escherichia coli infection in neonatal piglets. *Veterinary research*. 2018. Vol. 49, No. 1. P. 20. DOI: 10.1186/s13567-018-0511-4

23. Inoue R., Tsukahara T. Composition and physiological functions of the porcine colostrum. *Animal science journal = Nihon chikusan Gakkaiho*. 2021. Vol. 92, No. 1. P. e13618. DOI: 10.1111/asj.13618

24. Кокарєв А. В., Масюк Д. М. Формування клітинних механізмів імунного захисту у поросят за дії препарату «Імунолак». *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С. З. Ґжицького. Серія «Ветеринарні науки».* 2017. Т. 19. № 77. С. 214–219.

25. Кокарєв А.В. Формування фагоцитарної ланки імунітету поросят у ранньому постнатальному онтогенезі та її корекція препаратом «Імунолак» у ланцюзі мати-плід-новонароджений. *Збірник наукових праць Харківської державної зооветеринарної академії «Проблеми зооінженерії та ветеринарної медицини».* Харків, 2015. Вип. 31, Ч. 2. С. 89–94.

26. Krogh U., Quesnel H., Le Floch N., Simongiovanni A., van Milgen J. A dynamic mammary gland model describing colostrum immunoglobulin transfer and milk production in lactating sows. *Journal of animal science*. 2021. Vol. 99, No. 2. P. skab030. DOI: 10.1093/jas/skab030

27. Kulkarni D. H., Newberry R. D. Intestinal Macromolecular Transport Supporting Adaptive Immunity. *Cellular and molecular gastroenterology and hepatology.* 2019. Vol. 7, No. 4. P. 729–737. DOI: 10.1016/j.jcmgh.2019.01.003

28. Le Huërou-Luron I., Bouzerzour K., Ferret-Bernard S., Ménard O., Le Normand L., Perrier C., Le Bourgot C., Jardin J., Bourlieu C., Carton T., Le Ruyet P., Cuinet I., Bonhomme C., Dupont D. A mixture of milk and vegetable lipids in infant formula changes gut digestion, mucosal immunity and microbiota composition in neonatal piglets. *European journal of nutrition*. 2018. Vol. 57, No. 2. P. 463–476. DOI: 10.1007/s00394-016-1329-3

29. Lerch F., Vötterl J. C., Schwartz-Zimmermann H. E., Sassu E. L., Schwarz L., Renzhammer R., Bünger M., Sharma S., Koger S., Sener-Aydemir A., Quijada N. M., Selberherr E., Kummer S., Berthiller F., U Metzler-Zebeli B. Exposure to plant-oriented microbiome altered jejunal and colonic innate immune response and barrier function more strongly in suckling than in weaned piglets. *Journal of animal science.* 2022. Vol. 100, No. 11. P. skac310. DOI: 10.1093/jas/skac310

30. Lipsit S., Facciuolo A., Scruten E., Griebel P., Napper S. Plasma Cytokines and Birth Weight as Biomarkers of Vaccine-Induced Humoral Responses in Piglets. *Frontiers in veterinary science*. 2022. Vol. 9. P. 922992. DOI: 10.3389/fvets.2022.922992

31. Maciag S. S., Bellaver F. V., Bombassaro G., Haach V., Morés M. A. Z., Baron L. F., Coldebella A., Bastos A. P. On the influence of the source of porcine colostrum in the development of early immune ontogeny in piglets. *Scientific reports*. 2022. Vol. 12, No. 1. P. 15630. DOI: 10.1038/s41598-022-20082-1

32. Madsen P. A., Etheve S., Heegaard P. M. H., Skovgaard K., Mary A. L., Litta G., Lauridsen C. Influence of vitamin D metabolites on vitamin D status, immunity and gut health of piglets. *Veterinary immunology and immunopathology*. 2023. Vol. 257. P. 110557. DOI: 10.1016/j.vetimm.2023.110557

33. Martinez C. A., Rubér M., Rodriguez-Martinez H., Alvarez-Rodriguez M. Pig Pregnancies after Transfer of Allogeneic Embryos Show a Dysregulated Endometrial/Placental Cytokine Balance: A Novel Clue for Embryo Death? *Biomolecules.* 2020. Vol. 10, No. 4. P. 554. DOI: 10.3390/biom10040554

34. Masiuk D. M., Kokariev A. V., Bal R., Nedzvetsky V. S. The isotonic protein mixture suppresses Porcine Epidemic Diarrhea Virus excretion and initiates intestinal defensive response. *Theoretical and Applied Veterinary Medicine.* 2022. Vol. 10, No. 2. P. 23–28. https://doi.org/10.32819/2022.10009

35. Masiuk, D., Nedzvetsky, V., & Kokariev, A. (2023). Features of the functioning of the natural defense mechanisms of piglets under the influence of immunotropic substances. *Scientific Messenger of LNU of Veterinary Medicine and Biotechnologies. Series : Veterinary Sciences*, Vol. 25, No. 112. P. 181–192. https://doi.org/10.32718/nvlvet11229.

36. Merlot E., Pastorelli H., Prunier A., Père M. C., Louveau I., Lefaucheur L., Perruchot M. H., Meunier-Salaün M. C., Gardan-Salmon D., Gondret F., Quesnel H. Sow environment during gestation: part I. Influence on maternal physiology and lacteal secretions in relation with neonatal

survival. *Animal.* 2019. Vol. 13, Suppl 7. P. 1432–1439. DOI: 10.1017/S1751731118002987

37. Orije M. R. P., Maertens K., Corbière V., Wanlapakorn N., Van Damme P., Leuridan E., Mascart F. The effect of maternal antibodies on the cellular immune response after infant vaccination: A review. *Vaccine*. 2020. Vol. 38, No. 1. P. 20–28. DOI: 10.1016/j.vaccine.2019.10.025

38. Pietrasina O., Miller J., Rząsa A. Differences in the relative counts of peripheral blood lymphocyte subsets in various age groups of pigs. *Can J Vet Res.* 2020 Vol. 84, No. 1. P. 52–59.

39. Qi M., Tan B., Wang J., Liao S., Li J., Cui Z., Shao Y., Ji P., Yin Y. Postnatal growth retardation is associated with deteriorated intestinal mucosal barrier function using a porcine model. *Journal of cellular physiology*. 2021. Vol. 236, No. 4. P. 2631–2648. DOI: 10.1002/jcp.30028

40. Ray A., Bhati T., Arora R., Rastogi S. Progesterone-mediated immunoregulation of cytokine signaling by miRNA-133a and 101-3p in Chlamydia trachomatis-associated recurrent spontaneous abortion. *Molecular immunology*. 2023. Vol. 164. P. 47–57. DOI: 10.1016/j.molimm.2023.10.012

41. Schlosser-Brandenburg J., Ebner F., Klopfleisch R., Kühl A. A., Zentek J., Pieper R., Hartmann S. Influence of Nutrition and Maternal Bonding on Postnatal Lung Development in the Newborn Pig. *Frontiers in immunology*. 2021. Vol. 12. P. 734153. DOI: 10.3389/fimmu.2021.734153

42. Sinkora M., Butler J. E. Progress in the use of swine in develo-mental immunology of B and T lymphocytes. *Developmental and comparative immunology.* 2016. Vol. 58. P. 1–17. DOI: 10.1016/j.dci.2015.12.003

43. Wang J., Li Y., Yuan H., Shi S., Zhang L., Yang G., Pang W., Gao L., Cai C., Chu G. Effects of Alginic Acid on the Porcine Granulosa Cells and Maturation of Porcine Oocytes. *Molecular nutrition & food research*. 2023. Vol. 67, No. 22. P. e2300130. DOI: 10.1002/mnfr.202300130

44. Єфімов В. Г., Ракитянський В. М. Показники клітинного імунітету поросят на дорощуванні за впливу гумату натрію, бурштинової кислоти і мікроелементів. *Науковий вісник Львівського національного університету ветеринарної медицини та біотехнологій імені С. З. Ґжицького.* 2015. Т. 17, № 3. С. 32–37.

45. Yefimov V., Kostiushkevych K., Rakytianskyi V. Influence of feed additive from peat on morphological and biochemical blood profile of piglets. *Veterinarija ir Zootechnika*. 2017. № 75. С. 17–21.

Information about the authors:
Masiuk Dmytro Mykolaiovych,
Doctor of Veterinary Sciences, Professor,
Head of the Department of Physiology, Animal Biochemistry and Laboratory Diagnostics,
Dnipro State Agrarian and Economic University
25, Serhii Efremov str., Dnipro, 49009, Ukraine

Nedzvetsky Victor Stanislavovych,
Doctor of Biological Sciences, Professor,
Vice-director of Scientific Research Center of Biosafety and Environmental Control Agro-Industrial Complex,
Dnipro State Agrarian and Economic University
25, Serhii Efremov str., Dnipro, 49009, Ukraine

Kokariev Andrii Viktorovych,
Candidate of Veterinary Sciences, Associate Professor,
Head of the Department of Immunochemical and Molecular Genetic Analysis of Scientific Research Center of Biosafety and Environmental Control Agro-Industrial Complex,
Dnipro State Agrarian and Economic University
25, Serhii Efremov str., Dnipro, 49009, Ukraine