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Effect of bacterial lysate immunomodulator on key hematological parameters and functional activity of piglets' immune system during post-weaning

Eduard F. Sadikhov[1], Sergei V. Fedotov[1], Ekaterina S. Demidova[2]

[1] Russian State Agrarian University – Moscow Timiryazev Agricultural Academy, ul. Timiryazevskaya, 49, Moscow 127434, Russia

[2] Moscow State Academy of Veterinary Medicine and Biotechnology – MVA by K. I. Skryabin, ul. Academika Skryabina, 23, Moscow 109472, Russia

ABSTRACT

Introduction. Post-weaning period in pig production is a critical phase that determines the herd's future productivity and health. Stress from dietary changes, regrouping, and altered microclimate leads to post-weaning syndrome. Its underlying mechanism is transient immunosuppression resulting from hyperactivity of the hypothalamic-pituitary-adrenal axis and increased cortisol levels. For this reason, it is essential to develop immunocorrective strategies during this period.

Objective. To assess impact of an oral polyvalent bacterial lysate immunomodulator (*Bordetella bronchiseptica*, *Haemophilus parasuis*, and *Streptococcus suis*) on hematological parameters and immune system activation of piglets.

Materials and methods. Sixty blood samples of piglets taken from April to July 2024 on a pig farm in the Moscow Oblast were tested. Laboratory tests were conducted at the Russian State Agrarian University – Moscow Timiryazev Agricultural Academy using a hematology analyzer. Statistical analysis was performed using Statistica v.13.0.

Results. In a three-phase experiment involving experimental and control groups of piglets ($n = 30$), a course-based approach to Immbaclys C use induced pronounced changes in hematological parameters. A statistically significant and reproducible trend toward activation of the immunity lymphocyte component was revealed. All parameters remained within the normal range, indicating no negative impact of the tested medication on the hematopoiesis.

Conclusion. Three courses of Immbaclys C (14 days per one course, with a 21-day interval) induced a sustained immunostimulatory effect in piglets, along with improved zootechnical parameters. Hematological analysis revealed no deviations from normal values, confirming good tolerability and safety profile of the medication.

Keywords: hematology, immunity, immunomodulators, Immbaclys C, lymphocytes, phagocytosis

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For correspondence: Sergei V. Fedotov, Dr. Sci. (Veterinary Medicine), Professor, Head of the Department of Veterinary Medicine, Russian State Agrarian University – Moscow Timiryazev Agricultural Academy, ul. Pasechnaya, 2, Moscow 127550, Russia, *serfv@mail.ru*

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Влияние иммуномодулирующего препарата на основе бактериального лизата на ключевые гематологические параметры и функциональную активность иммунной системы поросят в период доращивания

Э. Ф. Садыхов[1], С. В. Федотов[1], Е. С. Демидова[2]

[1] ФГБОУ ВО «Российский государственный аграрный университет – МСХА имени К. А. Тимирязева» (ФГБОУ ВО РГАУ – МСХА имени К. А. Тимирязева), ул. Тимирязевская, 49, г. Москва, 127434, Россия

[2] ФГБОУ ВО «Московская государственная академия ветеринарной медицины и биотехнологии – МВА имени К. И. Скрябина» (ФГБОУ ВО МГАВМиБ – МВА имени К. И. Скрябина), ул. Академика Скрябина, 23, г. Москва, 109472, Россия

РЕЗЮМЕ

Введение. В свиноводстве постотъемный период – критическая фаза, определяющая будущую продуктивность и здоровье поголовья. Стресс от смены рациона, перегруппировки и изменения микроклимата приводят к постотъемному синдрому. Его основа – временная иммуносупрессия

из-за гиперактивности гипоталамо-гипофизарно-надпочечниковой оси и повышенного уровня кортизола, который подавляет иммунитет и разрушает лимфоидные клетки, снижая устойчивость поросят к респираторным и кишечным инфекциям. По этой причине разработка средств для иммунокоррекции в этот период крайне важна.

Цель исследования. Оценить влияние перорального иммуномодулятора на основе поливалентного бактериального лизата (*Bordetella bronchiseptica*, *Haemophilus parasuis* и *Streptococcus suis*) на гематологические показатели и активацию иммунной системы поросят.

Материалы и методы. Исследовали 60 образцов крови поросят, взятых с апреля по июль 2024 г. на свинокомплексе в Московской области. Лабораторные исследования проводили в ФГБОУ ВО «Российский государственный аграрный университет – МСХА имени К. А. Тимирязева» на гематологическом анализаторе. Статистическую обработку выполняли с использованием программы Statistica v.13.0.

Результаты. Трехфазный эксперимент с опытной ($n = 30$) и контрольной ($n = 30$) группами поросят показал, что курсовое применение препарата «Иммбаклиз С» оказало выраженную динамику гематологических показателей. Выявлена статистически значимая и воспроизводимая тенденция к активации лимфоцитарного звена иммунитета. Все показатели оставались в пределах нормы, что указывает на отсутствие негативного влияния исследуемого препарата на кроветворение.

Заключение. Трехкратное курсовое введение препарата «Иммбаклиз С» поросятам по схеме (14 дней каждый курс с интервалом 21 день) индуцирует устойчивый иммуностимулирующий эффект, сопровождающийся улучшением зоотехнических параметров. Гематологический анализ не выявил отклонений, что подтверждает хорошую переносимость и безопасность препарата.

Ключевые слова: гематология, иммунитет, иммуномодуляторы, «Иммбаклиз С», лимфоциты, фагоцитоз

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Конфликт интересов: Федотов С. В. является членом редколлегии журнала «Ветеринария сегодня», но никакого отношения к решению опубликовать эту статью не имеет. Рукопись прошла принятую в журнале процедуру рецензирования. Авторы декларируют отсутствие явных и потенциальных конфликтов интересов, связанных с публикацией настоящей статьи.

Для корреспонденции: Федотов Сергей Васильевич, д-р вет. наук, профессор, заведующий кафедрой ветеринарной медицины ФГБОУ ВО РГАУ – МСХА имени К. А. Тимирязева, ул. Пасечная, 2, г. Москва, 127550, Россия, *serfv@mail.ru*

INTRODUCTION

The post-weaning period is a critical stage in commercial pig farming, determining subsequent productivity and health status of the herd. This period is characterized by high levels of stress, leading to transient immunodeficiency and increased susceptibility of pigs to respiratory and enteric infections [1]. The abrupt switch to plant-based feed, alterations in social hierarchy, along with transport- and temperature-induced stress collectively trigger the so-called post-weaning syndrome, when immune system suppression turns out to be a key element of pathogenesis [2]. This condition is mediated by hyperactivation of the hypothalamic-pituitary-adrenal axis and the adrenal cortex, leading to excessive production cortisol, which demonstrates direct lymphocytotoxic and pro-apoptotic activity. Clinically, this results in reduced resistance and increased susceptibility to infectious diseases, mainly of the respiratory and gastrointestinal tracts. Immune status correction at this time represents a pressing task. Improving productive health and stress resilience in commercial pig farming requires a deep understanding of how various dietary substances affect physiological and biochemical processes in the body [3].

Domestic pig farming in the context of import substitution and food security is characterized by steady growth and is associated with the need for intensive production. High-density housing technologies boost productivity but also increase the risk of infectious disease spread, which is traditionally managed by wide use of antimicrobials. Given the global policy on reducing feed antibiotics and combating antimicrobial resistance, development and application of alternative biologicals capable of modulating the body's natural resistance is becoming a priority [4, 5].

Immunomodulators derived from antigenic complexes of relevant pathogens are of particular scientific and practical interest. Such medications specifically stimulate a specific immune response without exerting selective pressure on the microbiota or contributing to resistance development [4]. Although effectiveness of some immunotropic products in enhancing survival and average daily weight gain in weaned piglets has been well documented [5, 6, 7], the detailed impact of their course administration on dynamics of key hematologic and immunologic parameters remains underexplored in the scientific literature.

The objective of this study is to provide a comprehensive assessment of how course-based administration of Immbaclys C immunomodulator affects various immunity components, as reflected by the hematological profile of piglets during the post-weaning.

According to the World Health Organization ATC classification, Immbaclys C (NITA-FARM, Russia is the right holder and marketing authorization holder) belongs to the group of immunotropic agents (immunomodulators). The medication consists of coated granules with a modified-release formulation for oral use. Each gram of the medication contains the following active ingredient:

protein-lipopolysaccharide antigen complex derived from the lysate of *Bordetella bronchiseptica*, *Haemophilus parasuis* и *Streptococcus suis* (10 mg); and the following excipients: sodium glutamate, D-mannitol, propyl gallate, macrogol cetostearyl ether (polyethylene glycol-25-cetostearyl ether), sucrose, povidone K-30, quinoline yellow (E104), and chalk.

To achieve the set objective, a systematic analysis was performed, encompassing both published scientific data and the findings from our own experiments. The task was to compare dynamic changes in key hematological parameters, i.e. white blood cell (WBC) differential, red blood cell (RBC) and platelet (PLT) lineage parameters, in animals of the experimental and control groups, as well as to evaluate the immunomodulatory effect of the tested medication.

MATERIALS AND METHODS

The experiments were carried out on a commercial pig farm Mashkino SPK (Kolomna Urban District, Moscow Oblast) during the spring-summer season of 2024 (April through July). For the purposes of the study, two groups of clinically healthy crossbred piglets (Landrace × Large White, females) were formed according to the principle of analogues, with 30 animals in each group (n = 60). The animals, aged between 22 and 113 days with a live weight ranging from 7 to 48 kg, were kept under standard conditions in a single production facility without being moved throughout the entire experiment.

Both groups were kept under the same conditions (zoohygienic microclimate parameters, diet, veterinary care, and lighting schedule) to prevent violations of feeding and housing hygiene [8, 9]. In addition to the basic diet, the piglets of the experimental group received the tested immunomodulator Immbaclys C. The immunomodulator was administered according to the manufacturer's instructions at a dosage of 0.6 g per 1 kg of the mixed feed daily. The control group received a standard diet without additives.

The experiment design included three courses of the medication use, each lasting for 14 days, with 21-day intervals between the courses. During each course, venous blood was sampled four times (maximum 10 mL per sampling, consistent with bioethical guidelines) at precisely specified time points: day 0 (baseline), day 8 (anticipated onset of the immune response), day 15 (peak effect), and day 22 (course completed).

Laboratory tests were performed at the Department of Veterinary Medicine of the Russian State Agrarian University – Moscow Timiryazev Agricultural Academy. Hematological parameters were measured in dynamics.

To eliminate systematic errors during the experiment, a blind method was used, in which personnel responsible for animal care and sample collection were unaware of the animals' group assignment. No vaccines or antibacterials were permitted during the experiment. All procedures were approved by the local Bioethics Committee and complied with the principles of humane treatment of laboratory animals [10].

The data was statistically processed using Statistica v.13.0 software package. To assess the significance of differences between groups at the observation time points, the nonparametric Mann – Whitney U-test and Fisher's exact test were used. Differences were considered statistically significant at $p \leq 0.05$.

The age period (22–113 days) was chosen due to the pronounced morphofunctional restructuring of the gastrointestinal tract and immune system of piglets during post-weaning, which is characterized by maturation of lymphoid tissue and formation of the intestinal barrier [11]. Administering immunomodulators during this ontogenesis stage facilitates the optimal maturation of innate resistance and increases the animals' ability to withstand opportunistic pathogens [12]. The hematological parameters assessed in this study are highly sensitive markers of both innate and adaptive immune activity following administration of the antigenic complexes. Administration of such medications activates the immune system in pigs [13].

Standardization of external factors (automated feeding, microclimate control, feed testing for mycotoxins [14, 15]) together with individual housing of animals minimized variability unrelated to the immunomodulator's effect and ensured reproducibility of conditions in compliance with contemporary veterinary research standards [16].

RESULTS AND DISCUSSION

To assess the immunomodulator's effect on replacement gilts, we compared data from the control and experimental groups. Individual animal health indicators served as the basis for calculating mean group values for the main hematological parameters. The results are systematized in tables, which made it possible to conduct a comparative intergroup analysis, as well as to assess dynamical changes during each course of application and during the entire experiment.

Hematological parameters were tested before the start and during the three courses of Immbaclys C use, which was administered to piglets in the experimental group in accordance with the manufacturer's recommendations. The test results are given in Tables 1–3.

During Course 1, piglet blood was tested for dynamic hematological changes in the experimental group (n = 30) on Days 0, 8, 15, and 22.

The most pronounced changes were observed in the white blood cell (WBC) differential. Thus, the absolute lymphocyte (LYM) count increased from (8.57 ± 0.44) to (11.05 ± 0.43) × 10^9/L by day 22, which was accompanied by an increase in their relative count from (61.27 ± 1.47) to (71.94 ± 1.23)%. The neutrophil (NEU) count, in contrast, decreased both in absolute terms – from (5.24 ± 0.31) to (4.05 ± 0.19) × 10^9/L, and as a percentage – from (37.55 ± 1.45) to (26.30 ± 0.97)%. The monocyte (MON) count remained stable and within the reference range.

These shifts in the WBC differential – an increase in LYM count and a decrease in NEUs – may indicate activation of the cellular component of the immune system in response to Immbaclys C. Lymphocytes, as key effector cells of adaptive immunity, play a central role in pathogen recognition and neutralization, as well as in immunological memory [17].

Red blood cell parameters (erythrocyte, hemoglobin, hematocrit count) remained within physiological reference intervals throughout the first course of administration, indicating that the immunomodulator had no adverse effect on the hematopoietic system [18]. A moderate increase in RBC count was observed by day 15 – (7.16 ± 0.08) × 10^{12}/L, but by the end of the course the value had returned to baseline – (6.63 ± 0.11) × 10^{12}/L.

Table 1
Mean hematological parameters of piglets in the experimental group ($n = 30$) receiving Immbaclys C and piglets in the control group ($n = 30$) during the first treatment course

Parameters	Group	Quantitative values at various time points			
		Day 0	Day 8	Day 15	Day 22
White blood cells (WBC), $\times 10^9$/L (reference interval 11.0–22.0)	Experimental	13.98 ± 0.62	14.73 ± 0.77	14.87 ± 0.66	14.93 ± 0.49
	Control	16.34 ± 0.46	13.88 ± 0.31	13.46 ± 0.31	13.05 ± 0.49
Lymphocytes (LYM), $\times 10^9$/L (reference interval 4.0–14.0)	Experimental	8.57 ± 0.44	10.75 ± 0.6	10.79 ± 0.49	11.05 ± 0.43
	Control	11.76 ± 0.34	11.16 ± 0.20	11.16 ± 0.20	11.09 ± 0.18
Monocytes (MON), $\times 10^9$/L (reference interval 0.0–2.2)	Experimental	0.15 ± 0.03	0.10 ± 0.01	0.09 ± 0.005	0.11 ± 0.01
	Control	0.08 ± 0.003	0.82 ± 0.62	0.20 ± 0.01	0.19 ± 0.01
Neutrophils (NEU), $\times 10^9$/L (reference interval 2.0–18.4)	Experimental	5.24 ± 0.31	3.89 ± 0.25	4.02 ± 0.23	4.05 ± 0.19
	Control	4.33 ± 0.10	4.84 ± 0.07	5.18 ± 0.08	4.65 ± 0.07
Lymphocytes, % (reference interval 35.0–64.0)	Experimental	61.27 ± 1.47	72.85 ± 1.21	72.60 ± 1.05	71.94 ± 1.23
	Control	63.52 ± 0.62	63.86 ± 0.61	60.36 ± 0.46	58.13 ± 0.33
Neutrophils, % (reference interval 20.0–80.0)	Experimental	37.55 ± 1.45	26.54 ± 1.21	26.77 ± 1.05	26.30 ± 0.97
	Control	25.77 ± 0.51	27.63 ± 0.21	27.30 ± 0.23	25.13 ± 0.50
Red blood cells (RBC), $\times 10^{12}$/L (reference interval 5.0–8.0)	Experimental	6.24 ± 0.08	6.58 ± 0.07	7.16 ± 0.08	6.63 ± 0.11
	Control	5.66 ± 0.08	5.57 ± 0.09	5.56 ± 0.06	5.34 ± 0.06
Hemoglobin (HGB), mmol/L (reference interval 6.2–9.9)	Experimental	5.68 ± 0.08	6.06 ± 0.07	6.13 ± 0.09	6.18 ± 0.09
	Control	6.20 ± 0.03	5.44 ± 0.06	5.46 ± 0.03	5.69 ± 0.08
Hematocrit (HCT), % (reference interval 32.0–50.0)	Experimental	34.92 ± 0.46	36.41 ± 0.43	38.91 ± 0.53	36.78 ± 0.46
	Control	34.91 ± 0.37	35.24 ± 0.23	35.50 ± 0.18	36.17 ± 0.09
Platelets (PLT), $\times 10^9$/L (reference interval 300–700)	Experimental	448.34 ± 31.70	431.14 ± 31.34	523.28 ± 27.64	491.55 ± 21.28
	Control	398.60 ± 17.18	455.23 ± 12.22	499.86 ± 15.59	475.58 ± 16.34

Platelets count also corresponded to the reference intervals. A temporary increase was observed on day 15, reaching (523.28 ± 27.64) × 10^9/L, a finding that may reflect a physiological reaction to introduction of the antigenic components. Nevertheless, this increase remained within the reference interval and was not associated with any clinical signs.

Therefore, the hematological assessment demonstrates that Immbaclys C is well tolerated and is capable of activating the LYM-mediated component of the immune system while preserving the homeostasis of other blood elements [19].

Piglets of the control group ($n = 30$) that did not receive Immbaclys C, were monitored for dynamic changes in hematological parameters during the first course of the experiment, i.e. on days 0, 8, 15, and 22.

In the control group, the total WBC count gradually decreased from (16.34 ± 0.46) to (13.05 ± 0.39) × 10^9/L by day 22. At the same time, the absolute LYM count remained relatively stable, ranging from (11.76 ± 0.34) to (11.09 ± 0.18) × 10^9/L, while their percentage decreased from (63.52 ± 0.62) to (58.13 ± 0.33)%. Absolute NEU counts fluctuated within a narrow range (4.33–5.18) × 10^9/L; however, their relative proportion increased by day 8 to (27.63 ± 0.21)%, which may be attributed to natural age-related dynamics or background effects of housing conditions.

Notably, an abnormally high MON count occurred on day 8 (0.82 ± 0.62) × 10^9/L, sharply contrasting with the other values (0.08–0.20) × 10^9/L. Given the large standard deviation (± 0.62), this finding most probably results from sporadic outliers within the sample rather than indicating a systemic pathological process, particularly since no clinical signs of inflammation were observed in the animals [20].

Red blood cell parameters remained within reference intervals; however, a tendency toward a decrease in RBC

Table 2
Mean hematological parameters of piglets in the experimental group ($n = 30$) receiving Immbaclys C and piglets in the control group ($n = 30$) during the second course

Parameters	Group	Quantitative values at various time points			
		Day 0	Day 8	Day 15	Day 22
White blood cells (WBC), × 10^9/L (reference interval 11.0–22.0)	Experimental	15.86 ± 0.75	15.65 ± 0.61	15.49 ± 0.83	15.92 ± 0.65
	Control	14.66 ± 0.33	13.57 ± 0.31	13.74 ± 0.29	14.48 ± 0.28
Lymphocytes (LYM), × 10^9/L (reference interval 4.0–14.0)	Experimental	10.94 ± 0.61	9.50 ± 0.38	9.09 ± 0.41	12.33 ± 0.48
	Control	10.43 ± 0.22	9.92 ± 0.12	9.45 ± 0.04	10.19 ± 0.15
Monocytes (MON), × 10^9/L (reference interval 0.0–2.2)	Experimental	0.12 ± 0.01	0.12 ± 0.01	0.10 ± 0.007	0.14 ± 0.01
	Control	0.15 ± 0.007	0.17 ± 0.006	0.19 ± 0.006	0.22 ± 0.004
Neutrophils (NEU), × 10^9/L (reference interval 2.0–18.4)	Experimental	4.91 ± 0.28	5.91 ± 0.42	6.19 ± 0.93	5.23 ± 0.25
	Control	5.10 ± 0.09	5.06 ± 0.07	5.44 ± 0.04	5.43 ± 0.05
Lymphocytes, % (reference interval 35.0–64.0)	Experimental	67.73 ± 1.49	62.29 ± 1.64	60.57 ± 1.91	69.59 ± 1.28
	Control	58.65 ± 0.70	58.37 ± 0.48	55.38 ± 0.40	57.49 ± 0.52
Neutrophils, % (reference interval 20.0–80.0)	Experimental	31.42 ± 1.51	38.31 ± 1.92	38.74 ± 1.85	32.69 ± 1.51
	Control	35.29 ± 0.75	35.60 ± 0.48	36.23 ± 0.25	36.30 ± 0.21
Red blood cells (RBC), × 10^{12}/L (reference interval 5.0–8.0)	Experimental	6.71 ± 0.08	6.74 ± 0.09	6.92 ± 0.10	6.77 ± 0.09
	Control	6.21 ± 0.12	6.11 ± 0.06	6.02 ± 0.04	5.90 ± 0.06
Hemoglobin (HGB), mmol/L (reference interval 6.2–9.9)	Experimental	5.85 ± 0.04	5.65 ± 0.08	5.71 ± 0.09	5.96 ± 0.07
	Control	6.29 ± 0.06	6.26 ± 0.03	6.31 ± 0.03	6.37 ± 0.04
Hematocrit (HCT), % (reference interval 32.0–50.0)	Experimental	38.28 ± 0.32	36.37 ± 0.47	36.66 ± 0.54	39.02 ± 0.50
	Control	38.23 ± 0.32	37.77 ± 0.52	37.77 ± 0.52	36.02 ± 0.30
Platelets (PLT), × 10^9/L (reference interval 300–700)	Experimental	524.92 ± 28.82	537.83 ± 26.62	615.59 ± 23.45	551.72 ± 18.63
	Control	441.00 ± 18.13	458.83 ± 16.76	473.07 ± 20.83	428.34 ± 10.91

count from (5.66 ± 0.08) to (5.34 ± 0.06) × 10^{12}/L, as well as in hemoglobin (HGB) level from (6.20 ± 0.03) to (5.69 ± 0.08) mmol/L, was noted by the end of the observation period.

Platelet counts were within the normal range. A moderate increase in PLT count was observed, from (398.6 ± 17.18) to (499.86 ± 15.59) × 10^9/L by day 15, followed by a decrease by the end of the period.

Accordingly, the hematological findings in the control group reflect normal ontogenetic development and show no evidence of pathological alterations.

During the second Immbaclys C course, blood was sampled from experimental piglets (n = 30) on days 0, 8, 15, and 22 for dynamic hematological analysis. All indicators remained within the physiological reference intervals typical for this age group [17].

The total WBC count remained stable throughout the second course, ranging from (15.49 ± 0.83) to (15.92 ± 0.65) × 10^9/L, reflecting a lack of systemic inflammation. Marked dynamics were observed in the WBC differential: the absolute LYM count decreased by day 15 to (9.09 ± 0.41) × 10^9/L, but by the end of the course it had increased significantly to (12.33 ± 0.48) × 10^9/L. Accordingly, the relative LYM count fluctuated from (60.57 ± 1.91) to (69.59 ± 1.28)%, remaining within the reference range (35.0–64.0%) except for the values on days 0 and 22, which slightly exceeded the upper reference limit. Simultaneously, an inverse change in NEU content was observed: their proportion increased by day 15 to (38.74 ± 1.85)% and decreased by the end of the course to (32.69 ± 1.51)%. Monocyte levels remained consistently minimal – (0.10–0.14) × 10^9/L – indicating the absence of chronic inflammation.

Red blood cell parameters remained consistently stable, RBC count ranged (6.71–6.92) × 10^{12}/L, HGB ranged from 5.65 to 5.96 mmol/L, and HCT ranged from 36.37%

Table 3
Mean hematological parameters of piglets in the experimental group ($n = 30$) receiving Immbaclys C and piglets in the control group ($n = 30$) during the third course

Parameters	Group	Quantitative values at various time points			
		Day 0	Day 8	Day 15	Day 22
White blood cells (WBC), $\times 10^9$/L (reference interval 11.0–22.0)	Experimental	14.69 ± 0.53	15.34 ± 0.44	15.91 ± 0.72	16.43 ± 0.41
	Control	13.26 ± 0.40	14.06 ± 0.29	13.77 ± 0.54	13.82 ± 0.35
Lymphocytes (LYM), $\times 10^9$/L (reference interval 4.0–14.0)	Experimental	11.02 ± 0.27	9.89 ± 0.24	11.67 ± 0.51	13.22 ± 0.49
	Control	11.21 ± 0.30	10.74 ± 0.20	12.66 ± 0.41	12.97 ± 1.07
Monocytes (MON), $\times 10^9$/L (reference interval 0.0–2.2)	Experimental	0.16 ± 0.01	0.13 ± 0.008	0.29 ± 0.07	0.14 ± 0.06
	Control	0.75 ± 0.54	0.16 ± 0.003	0.19 ± 0.005	0.18 ± 0.006
Neutrophils (NEU), $\times 10^9$/L (reference interval 2.0–18.4)	Experimental	5.40 ± 0.27	6.11 ± 0.28	5.43 ± 0.26	5.19 ± 0.17
	Control	5.35 ± 0.07	6.12 ± 0.12	6.36 ± 0.23	5.50 ± 0.05
Lymphocytes, % (reference interval 35.0–64.0)	Experimental	65.73 ± 1.43	60.35 ± 1.87	69.21 ± 1.41	71.35 ± 0.98
	Control	56.09 ± 0.76	57.45 ± 0.56	58.28 ± 0.49	56.61 ± 0.51
Neutrophils, % (reference interval 20.0–80.0)	Experimental	34.51 ± 1.66	38.83 ± 1.76	31.38 ± 1.18	32.58 ± 0.49
	Control	33.74 ± 0.85	36.66 ± 0.44	30.08 ± 0.58	34.04 ± 0.27
Red blood cells (RBC), $\times 10^{12}$/L (reference interval 5.0–8.0)	Experimental	6.67 ± 0.08	6.88 ± 0.10	6.53 ± 0.04	6.59 ± 0.06
	Control	6.47 ± 0.07	6.93 ± 0.08	6.53 ± 0.04	6.22 ± 0.12
Hemoglobin (HGB), mmol/L (reference interval 6.2–9.9)	Experimental	5.48 ± 0.06	5.74 ± 0.12	5.97 ± 0.05	6.12 ± 0.07
	Control	5.48 ± 0.05	5.67 ± 0.05	5.78 ± 0.02	5.78 ± 0.05
Hematocrit (HCT), % (reference interval 32.0–50.0)	Experimental	35.83 ± 0.47	37.14 ± 0.66	38.06 ± 0.30	38.62 ± 0.38
	Control	34.00 ± 0.27	36.92 ± 0.51	38.86 ± 0.51	38.47 ± 0.75
Platelets (PLT), $\times 10^9$/L (reference interval 300–700)	Experimental	521.26 ± 21.84	529.39 ± 16.43	513.37 ± 21.50	467.86 ± 15.78
	Control	481.96 ± 9.98	532.20 ± 16.37	435.77 ± 13.42	449.17 ± 10.96

to 39.02%. This finding reflects the physiological heterogeneity of the erythrocyte population, which is attributable to normal erythropoiesis.

Platelet parameters also remained within the normal range. A moderate increase in PLT count was observed by day 15, reaching (615.59 ± 23.45) × 10^9/L, indicating normal functional activity of the megakaryocytic lineage.

Thus, during the second course of Immbaclys C no signs of toxic or pathological effects on the hematopoietic system were detected in piglets. The observed changes in the WBC differential, particularly the increase in LYM count by the end of the course, may indicate reactivation of the cellular component of the immune system in response to the antigenic components of the medication, which is consistent with its claimed immunomodulatory effect [21].

Dynamic hematological monitoring performed during the same corresponding period in the control group of piglets ($n = 30$) on days 0, 8, 15, and 22 showed that total WBC count fluctuated within a narrow range from (13.57 ± 0.31) to (14.66 ± 0.33) × 10^9/L, indicating the absence of a systemic inflammatory or stress response. The WBC differential showed a LYM predominance, with absolute LYM counts ranging between 9.45 and 10.43 × 10^9/L and relative percentages ranging from 55.38 to 58.65% – values consistent with the age-related reference range for piglets during the post-weaning period. Neutrophils accounted for 35.29–36.30% and, like monocytes, remained within the reference ranges, indicating a stable functional state of innate immunity.

Red blood cell parameters showed a gradual decrease in RBC count from (6.21 ± 0.10) to (5.90 ± 0.06) × 10^{12}/L, while HGB levels remained stable and even increased slightly from (6.29 ± 0.06) to (6.37 ± 0.04) mmol/L. Hematocrit levels fell from (38.23 ± 0.32) to (36.02 ± 0.30)% – a change

that likely reflects physiological hemodilution associated with rapid growth and expansion of circulating plasma volume [22, 23, 24].

Platelet counts were also within the reference range. Their count ranged from (428.34 ± 10.91) to (473.07 ± 20.83) × 10^9/L.

Accordingly, the hematological findings in the control group throughout the second treatment course demonstrate normal ontogenetic development and absence of pathological changes [25], thereby establishing a robust reference for assessing Immbaclys C effects in the experimental group.

Throughout the third treatment course with Immbaclys C, serial hematological assessments were carried out in experimental piglets (n = 30) on days 0, 8, 15, and 22. All measured values remained within the normal physiological limits for 3–4-month-old piglets [26, 27].

The total WBC count showed a gradual increase from (14.69 ± 0.53) to (16.43 ± 0.41) × 10^9/L, indicating sustained immune system activity. The most pronounced changes were observed in the WBC differential. The relative LYM count progressively increased from (65.73 ± 1.43) to (71.35 ± 0.98)% by the end of the course, exceeding the upper limit of the reference range (64.0%). The absolute LYM count also increased from (11.02 ± 0.27) to (13.22 ± 0.49) × 10^9/L. Concurrently, the NEU percentage declined from (34.51 ± 1.66) to (32.58 ± 0.49)%, although absolute NEU counts stayed within the reference range. Such a shift of the WBC differential toward lymphocytosis indicates sustained activation of the cellular component of adaptive immunity, which is consistent with the mechanism of action of a medication containing antigenic components of *Bordetella bronchiseptica*, *Haemophilus parasuis*, and *Streptococcus suis*.

Red blood cell parameters remained stable; RBC count fluctuated within the range of (6.53–6.88) × 10^{12}/L, HGB ranged from 5.48 to 6.12 mmol/L, and HCT from 35.83 to 38.62%.

Platelet parameters remained within the normal range; PLT count varied from (467.86 ± 15.78) to (529.39 ± 16.43) × 10^9/L, indicating normal physiological PLT activity [28].

Thus, during the third course of Immbaclys C no signs of toxic or pathological effects on the hematopoietic system were detected in piglets. The observed progressive increase in LYMs confirms persistence and enhancement of the immunomodulatory effect of the medicinal product even with repeated course administration, indicating its high biological activity and good tolerability.

During the period corresponding to the third course of the experiment, dynamic hematological monitoring was also performed in the control group of piglets (n = 30) that did not receive Immbaclys C on days 0, 8, 15, and 22.

The total WBC count fluctuated within a narrow range, from (13.26 ± 0.40) to (14.06 ± 0.29) × 10^9/L, indicating the absence of a systemic inflammatory or stress response. The WBC differential showed LYM predominance, with relative LYM counts ranging from 56.09 to 58.28% – values that fall within the expected age-related reference range for piglets during the post-weaning. The absolute LYM count ranged from 10.74 to 12.97 × 10^9/L, with a significant increase observed on days 15 and 22, which may be attributed to natural maturation of the immune system. Neutrophils accounted for 30.08–36.66%, which is also within the reference ranges.

It should be noted that an abnormally high MON count was observed on day 0, equal to (0.75 ± 0.54) × 10^9/L, which is likely due to sporadic outliers in the sample, since subsequent values stabilized at (0.16–0.19) × 10^9/L and were not accompanied by clinical signs of inflammation.

Red blood cell parameters remained consistently stable; RBC count ranged from 6.22 to 6.93 × 10^{12}/L, HGB ranged from 5.48 to 5.78 mmol/L, and HCT ranged from 34.00 to 38.86%.

Platelet count also corresponded to the reference values and ranged from (435.77 ± 13.42) to (532.20 ± 16.37) × 10^9/L, which confirms their normal physiological activity.

Accordingly, the hematological findings in the control group throughout the third treatment course demonstrate normal ontogenetic development and absence of pathological changes, thereby establishing a robust reference for assessing Immbaclys C effects in the experimental group.

CONCLUSION

The three-phase experimental design made it possible to assess dynamics of hematological parameters in piglets of the experimental group (n = 30) during course administration of immunomodulator Immbaclys C and to compare them with the parameters of the control group (n = 30) that did not receive the medication. Analysis of data from the first, second, and third courses revealed a clear, reproducible, and progressive trend – a gradual strengthening of the LYM component of the immune system while all parameters remained within physiological norms.

Lymphocyte response dynamics. On day 22 of each course, the relative LYM count in the experimental group was as follows:

– course 1: (71.94 ± 1.23)%;
– course 2: (69.59 ± 1.28)%;
– course 3: (71.35 ± 0.98)%.

All values exceeded the upper limit of the reference range (64.0%) but remained stable and were not accompanied by disease clinical signs. In contrast, LYM percentages in the control group at the corresponding time points remained below 58.13; 57.49, and 56.61%, respectively, with statistically significant differences observed at all comparison points ($p \leq 0.05$).

The absolute LYM count also demonstrated a sustained increase: from (11.05 ± 0.43) × 10^9/L at the end of the first course to (13.22 ± 0.49) × 10^9/L on day 22 of the third course, whereas in the control group, greater variability and no clear trend were observed.

This dynamics demonstrated formation of a specific adaptive immune response to the antigenic components of the medication (*Bordetella bronchiseptica*, *Haemophilus parasuis*, and *Streptococcus suis*) and confirmed its claimed immunomodulatory effect.

General state of the hematopoietic system. RBC and PLT lineage parameters remained within the age-specific norm throughout all three courses in both the experimental and control groups. Minor fluctuations in certain blood parameters corresponded to the physiological characteristics of intensive growth in piglets aged 1–4 months and had no pathological significance.

Importantly, no mortality was observed in either group. All piglets exhibited normal appetite, behavioral activity, and growth rates that were comparable to those

of the control animals. This confirms the absence of toxic, allergic, or stress-induced effects of the medication even after three courses of administration.

Specificity of the immune response. The decrease in relative NEU count in the experimental group (down to 26–33% vs. 34–36% in the control) accompanied by an increase in LYMs indicates a shift of the immune response toward cellular immunity, which is a desirable effect in prevention of bacterial infections caused by intracellular and opportunistic pathogens.

Oral administration of the polyvalent bacterial lysate-based immunomodulator during post-weaning induces statistically significant changes in the hematological profile of piglets, characterized by persistent leukocytosis due to absolute lymphocytosis while maintaining the neutrophil-to-lymphocyte ratio, indicating immunostimulation without exacerbating stress burden.

The three-stage comparative analysis of hematological parameters allows us to draw the following conclusions:

– the course-based administration of Immbaclys C induces a specific and reproducible immunomodulatory response, manifested as a statistically significant and progressive increase in the relative and absolute LYM counts in peripheral blood;

– observed shift in the WBC differential (lymphocytosis accompanied by a decrease in NEU proportion) indicates a balance shift toward activation of adaptive cellular immunity, which is the targeted pharmacodynamic effect of the medication;

– the absence of significant deviations in RBC and PLT lineage parameters, together with the preserved clinical health, confirms safety and good tolerability of Immbaclys C even after repeated treatment course;

– the obtained findings support using this immunomodulator in prevention protocol for bacterial infections induced by antigenically related microorganisms, through selective stimulation of key effector mechanisms of specific immune defense;

– the obtained findings support using the bacterial lysate-based medications in veterinary preventive protocols during post-weaning to reduce infectious morbidity and reduce antibiotic usage in pig farming.

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INFORMATION ABOUT THE AUTHORS / ИНФОРМАЦИЯ ОБ АВТОРАХ

Eduard F. Sadikhov, Postgraduate Student, Department of Veterinary Medicine, Russian State Agrarian University – Moscow Timiryazev Agricultural Academy, Moscow, Russia; *eveterinar@mail.ru*

Sergei V. Fedotov, Dr. Sci. (Veterinary Medicine), Professor, Head of the Department of Veterinary Medicine, Russian State Agrarian University – Moscow Timiryazev Agricultural Academy, Moscow, Russia; *https://orcid.org/0000-0002-0004-3639, serfv@mail.ru*

Ekaterina S. Demidova, Cand. Sci. (Biology), Associate Professor, Department of Advanced Technologies in Poultry Farming, Moscow State Academy of Veterinary Medicine and Biotechnology – MVA by K. I. Skryabin, Moscow, Russia; *mixalysha@mail.ru*

Садыхов Эдуард Фамилович, аспирант кафедры ветеринарной медицины ФГБОУ ВО РГАУ – МСХА имени К. А. Тимирязева, г. Москва, Россия; *eveterinar@mail.ru*

Федотов Сергей Васильевич, д-р вет. наук, профессор, заведующий кафедрой ветеринарной медицины ФГБОУ ВО РГАУ – МСХА имени К. А. Тимирязева, г. Москва, Россия; *https://orcid.org/0000-0002-0004-3639, serfv@mail.ru*

Демидова Екатерина Сергеевна, канд. биол. наук, доцент кафедры передовых технологий в птицеводстве ФГБОУ ВО МГАВМиБ – МВА имени К. И. Скрябина, г. Москва, Россия; *mixalysha@mail.ru*

Contribution of the authors: Sadikhov E. F., Demidova E. S. – performing information retrieval and analysis, research concept, analysis of laboratory research results, compilation of tables, writing the manuscript; Fedotov S. V. – academic supervision, manuscript editing.

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