# УКРАЇНСЬКИЙ ЧАСОПИС ВЕТЕРИНАРНИХ НАУК

**Видавець:**
Національний університет біоресурсів і природокористування України

**Рік заснування: 2010**

*Рекомендовано до друку та поширення*
*через мережу Інтернет Вченою радою*
*Національного університету біоресурсів і природокористування України*
*(протокол № 8 від 27 лютого 2025 р.)*

**Рішення Національної ради України**
**з питань телебачення і радіомовлення № 1795,**
протокол № 31 від 21.12.2023 р.
Ідентифікатор медіа - R30-02293.

**Журнал входить до переліку наукових фахових видань України**

Категорія «Б». Галузь наук – Ветеринарні,
спеціальність – 211 «Ветеринарна медицина»
(Наказ Міністерства освіти і науки України від 28 грудня 2019 р. № 1643)

**Журнал представлено у міжнародних наукометричних базах даних, репозитаріях та пошукових системах**: Google Scholar, Національна бібліотека України імені В. І. Вернадського, BASE, Directory of Open Access Journals (DOAJ), CAB Abstracts and Global Health (CABI), Polska Bibliografia Naukowa (PBN), Ulrichsweb, Dimensions, University of Oslo Library, University of Hull Library, SOLO – Search Oxford Libraries Online, European University Institute, Leipzig University Library, Cambridge University Library, UCSB Library, German Union Catalogue of Serials, OUCI (Open Ukrainian Citation Index), WorldCat, Фахові видання України, EBSCO, Litmaps

**Адреса редакції**:
Національний університет біоресурсів і природокористування України
03041, вул. Героїв Оборони, 15, м. Київ, Україна
E-mail: info@veterinaryscience.com.ua
https://veterinaryscience.com.ua/uk

# UKRAINIAN JOURNAL OF VETERINARY SCIENCES

**Publisher:**
National University of Life and Environmental Sciences of Ukraine

**Year of foundation: 2010**

*Recommended for printing and distribution*
*via the Internet by the Academic Council*
*of National University of Life and Environmental Sciences of Ukraine*
*(Minutes No. 8 of February 27, 2025)*

**Decision of the National Council**
**of Television and Radio Broadcasting of Ukraine No. 1795,**
Minutes No. 31, dated 21.12.2023.
Media identifier - R30-02293.

**The journal is included in the list of professional publications of Ukraine**
Category "B". 0841 – Veterinary
(Order of the Ministry of Education and Science of Ukraine
of December 28, 2019, No. 1643)

**The journal is presented international scientometric databases, repositories and scientific systems**: Google Scholar, Vernadsky National Library of Ukraine, BASE, Directory of Open Access Journals (DOAJ), CAB Abstracts and Global Health (CABI), Polska Bibliografia Naukowa (PBN), Ulrichsweb, Dimensions, University of Oslo Library, University of Hull Library, SOLO - Search Oxford Libraries Online, European University Institute, Leipzig University Library, Cambridge University Library, UCSB Library, German Union Catalogue of Serials, OUCI (Open Ukrainian Citation Index), WorldCat, Professional publications of Ukraine, EBSCO, Litmaps

**Editors office address**:
National University of Life and Environmental Sciences of Ukraine
03041, 15 Heroiv Oborony Str., Kyiv, Ukraine
E-mail: info@veterinaryscience.com.ua
https://veterinaryscience.com.ua/en

UKRAINIAN JOURNAL OF VETERINARY SCIENCES

**2025. Volume 16, No. 1. P. 9-29**

Received: 16.11.2024 Revised: 24.01.2025 Accepted: 27.02.2025

DOI: 10.31548/veterinary1.2025.09

UDC: 636:001.891.53:615

# The influence of a prebiotic on the development of laboratory animals

**Jędrzej Jaskowski**
Doctor of Habilitation, Professor
Nicolaus Copernicus University
87100, 11 Jurij Gagarin Str., Toruń, Poland
https://orcid.org/0000-0002-3274-8692

**Vasyl Lyasota**
Doctor of Veterinary Sciences, Professor
Bila Tserkva National Agrarian University
09117, 8/1 Soborna Sq., Bila Tserkva, Ukraine
https://orcid.org/0000-0002-2442-2174

**Svitlana Tkachuk**
Doctor of Veterinary Sciences, Professor
National University of Life and Environmental Sciences of Ukraine
03041, 15 Heroiv Oborony Str., Kyiv, Ukraine
https://orcid.org/0000-0002-6923-1793

**Nadiia Bogatko**
Doctor of Veterinary Sciences, Professor
Bila Tserkva National Agrarian University
09117, 8/1 Soborna Sq., Bila Tserkva, Ukraine
https://orcid.org/0000-0002-1566-1026

**Aliona Bogatko**
Assistant
Bila Tserkva National Agrarian University
09117, 8/1 Soborna Sq., Bila Tserkva, Ukraine
https://orcid.org/0000-0001-8089-5884

**Abstract.** Prebiotics contribute to improved animal health; however, their beneficial effects on the body depend on the composition and dosage of the preparation, as well as the age and species of the animals. This study aimed to examine the effects of a new complex prebiotic, Bio-active,

***Suggested Citation:***
Jaskowski, J., Lyasota, V., Tkachuk, S., Bogatko, N., & Bogatko, A. (2025). The influence of a prebiotic on the development of laboratory animals. *Ukrainian Journal of Veterinary Sciences*, 16(1), 9-29. doi: 10.31548/veterinary1.2025.09.

*Corresponding author

on the physiology of white mice and rabbits. The experiment involved 40 white laboratory mice and rabbits. The prebiotic was administered at a dosage of 1.2 g per animal. Standard methods were used to assess the microclimate, the quality of tap water used for drinking, growth dynamics, haematological parameters, and microscopic examination of caecal mucosa smears (in rabbits). The microclimate parameters and water quality indicators met the requirements of current regulatory standards. Administration of the prebiotic to white laboratory mice resulted in a gradual increase in body weight and average daily weight gain throughout the study period. In rabbits, body weight, absolute and relative growth rates increased, contributing to improved survival rates and growth performance. It was demonstrated that the inclusion of the prebiotic in animal feed led to higher red blood cell counts, increased haemoglobin concentration, and elevated levels of total protein and globulins. The prebiotic also influenced the quantity and ratio of Gram-positive and Gram-negative microorganisms in the caecum of rabbits. Specifically, the number of Gram-negative microorganisms decreased by 17.7% ($P < 0.05$), while Gram-positive microorganisms increased by 19.4% ($P < 0.05$). The pH of the caecal content in the experimental group was 6.5, compared to 7.1 in the control group, indicating the restoration of functional capacity in the large intestine. Based on the results obtained, the components of the prebiotic at a dosage of 1.2 g per animal were found to have a positive effect on body weight gain, haematological parameters, and the quantitative composition of the large intestinal microbiota. These outcomes are significant for rearing healthy livestock and producing high-quality food products

**Keywords**: white mice; rabbits; housing conditions; growth performance; haematological parameters; microorganisms; caecum

## Introduction

A relevant strategy in contemporary animal husbandry is the utilisation of promising alternative immunostimulants, which can help to reduce the overuse of antibiotics in livestock farming. Adopting this approach addresses a key aim of the One Health concept, which is to prevent the spread of dangerous zoonotic diseases, produce safe and high-quality food products, and adhere to animal welfare standards.

J. Jing *et al.* (2023) pointed out that prebiotics are nutrients beneficial to human health that are fermented by microorganisms, leading to specific alterations in their composition. In essence, prebiotics are substances that nourish the gut microbiome, thereby regulating its population to favour the production of postbiotics and facilitating the removal of harmful metabolites and exogenous substances. Consequently, for the animal body, particularly during periods of growth and development or when certain diseases arise, the inclusion of prebiotic supplements in the basic diet is essential. The authors concluded that the ongoing development of novel research methods, such as sequencing and artificial intelligence, alongside a more thorough investigation into the mechanisms of action of probiotics, prebiotics, and postbiotics, will further their application in relevant sectors of animal product production.

The application of prebiotics in animal husbandry and the production of high-quality, safe food is crucial for human health. In this context, A. Ballini *et al.* (2023) and V.O. Oluwatobi *et al.* (2024) established that the human gut microbiota contains over 500 species of bacteria and constitutes 95% of the total number of cells in

the body. The authors highlighted that disruptions in the composition of the gut microbiota, resulting from poor diet, obesity, and a sedentary lifestyle, lead to dysfunction in numerous organs and systems within the body. A consequence of such disturbances is a reduction in the body's overall resistance to infectious diseases.

Given the proven beneficial effects of prebiotics, they are now widely used in the rearing of productive livestock (Shkromada *et al*., 2024). Developers of new formulations are particularly focused on the component composition, dosage, and in-depth investigations into the effects of both individual components and their combined action. For instance, amino acids are frequently included in the composition of novel prebiotic preparations, as the process of amino acid catabolism is vital for controlling the metabolism of various biological processes and regulating their mechanisms. In this regard, M. Beaumont *et al.* (2022) demonstrated that amino acids can modulate the composition of the gut microbiota, which influences the oxidation of amino acids and the production of metabolites in the intestine. The ability of bacteria to generate amino acid metabolites in the gut promotes the growth of beneficial microorganisms that can outcompete pathogenic ones. Consequently, research in this area has led to the introduction of the term "aminobiotics", which function as prebiotics within the body.

I. Ishii & M. Bhatia (2023) noted that in the human and animal body, proteins are synthesised from at least 20 of the hundreds of amino acids found in nature. Of particular importance are nine essential amino acids, which are not synthesised by the body's cells and must be obtained through diet. The authors highlighted polymers composed of a specific type of amino acid, known as an α-amino acid, which forms proteins.

The composition of prebiotics can also include live microorganism cultures. Consequently, the role of probiotic cultures in regulating animal and human health is continuing to evolve. D. Guzmán-Escalera *et al*. (2025) indicated that the action of metabolites from lactic acid fermentation leads to an increase in short-chain fatty acids, resulting in an anti-inflammatory effect. Furthermore, the action of bioactive peptides, also metabolites of lactic acid fermentation, leads to antihypertensive, antithrombotic, antioxidant, and antimicrobial effects. Additionally, L.L.C. De Jesus *et al.* (2024) identified several plasmids in *L. delbrueckii* that reduce the spread of antibiotic resistance genes, and this type of bioactive peptide metabolite from lactic acid fermentation has a lower propensity to act as a pathogen.

This research aimed to evaluate the impact of the prebiotic Bio-active at a dosage of 1.2 g per animal on the growth and development of white mice and rabbits. This will involve determining the changes in body weight, internal organs, haematological parameters, and the quantitative composition of the caecal microflora in rabbits.

## Literature Review

A primary requirement for the component composition of feed supplements for productive livestock, particularly rabbits, remains the inclusion of natural substances that can serve as alternatives to synthetic drugs. K. El-Sabrout *et al.* (2023) found that rabbit farming is currently facing a feed deficit due to intense competition within the animal husbandry sector, climate change, and wartime conditions. Consequently, the development and application of natural feed additives as substitutes for antibiotics in rabbit nutrition is of critical importance for raising healthy animal populations.

When administering prebiotics with varying component compositions to productive animals, investigating the state of the intestine is a priority. It is well-established that the intestine is one of the most vital organs, responsible for

the absorption of water and nutrients essential for maintaining the physiological functions of the animal body. A key function of the intestine is its barrier role, which is necessary for the elimination of harmful substances such as toxins, allergens, and pathogens. This was highlighted by K. Kobayashi *et al.* (2023), who demonstrated using an epithelial model of the small intestine that both live and heat-treated *L. bulgaricus* 2038 and *S. thermophilus* 1131 improved the condition of the intestine in cases of intestinal barrier dysfunction by modulating the integrity of the apical tight junction (TJ) protein in Caco-2 cells.

J. Jing *et al.* (2023), based on the findings of their research, elucidated the key mechanisms and positive impacts of prebiotics used to restore the homeostasis of the gut microbiota. Y. Zhang *et al.* (2023) noted that the functional capacity of the gut microbiome influences the extent to which nutrients from feed are absorbed, the development or progression of various diseases, and the body's ability to maintain protective functions against opportunistic and pathogenic microorganisms. Similarly, Y.P. Silva *et al.* (2020) demonstrated that the gut microbiome affects the functioning of organs and systems within the body, such as the nervous and immune systems, by acting through microbial signalling and metabolites, including short-chain fatty acids (SCFAs). Research by W. He *et al.* (2024) has shown that feeding mice with lyophilised powder made from fermented whey can regulate the quantity and ratio of the gut microbiota and improve the condition of the experimentally damaged nervous system in mice. This is attributed to the presence of surface receptors for neurotransmitters in the taxa that constitute the gut flora. Consequently, the functions of the central nervous system are closely interconnected with the activity of the intestine.

The digestion of feed and detoxification of substances within the intestine is possible through the action of specific types of microflora present in quantities that ensure an optimal balance in the large intestine. I.A. Biben *et al.* (2021) reached this conclusion and established that the species composition of the gut microflora influences the properties of lymphoid tissue, which is predominantly located in the large intestine. V.V. Kunovsky *et al.* (2024) established that commensal microorganisms regulate the connection between the human gut microbiota and the central nervous system. The gut-brain axis (GBA) comprises a bidirectional communication pathway between the central and enteric nervous systems, linking the emotional and cognitive centres of the brain with peripheral intestinal functions. Furthermore, S.A. Burmei & N.B. Boyko (2024) demonstrated that probiotic preparations should not exhibit any inhibitory effects on commensal microorganisms, and various types of biopreparations (pre-, pro-, syn-, and metabiotics) are employed to restore and correct the microbiota of the digestive tract. This approach forms the foundation for the development of nextgeneration probiotics, which promises to be a significant advancement in the production of new-generation biopreparations.

Vitamins A, E, and B complex are important constituents of prebiotic preparations. M. Asadi *et al.* (2024) highlighted the wide range of metabolic pathways initiated in the animal body through the use of B vitamins, which act as cofactors. When vitamin B complex was administered via injection to pregnant goats, a reduction in the incidence of metabolic diseases was observed, along with a significant increase ($P < 0.05$) in the red blood cell count, haemoglobin concentration, haematocrit level, and overall antioxidant status of their offspring.

Y. Shastak & W. Pelletier (2024) demonstrated that the fat-soluble vitamins A and E play a crucial and interconnected role in various biological processes. They also noted that a deficiency in these vitamins (hypovitaminosis) can lead to severe consequences for the animal

body, including impaired vision, reduced immunity, stunted growth, decreased reproductive capacity, and the development of nervous system disorders. The authors concluded that feed supplements containing vitamins A and E in innovative liquid formulations can prevent the development of diseases and improve the general health and productivity of animals.

M. Koprivica & A. Miljković (2024) investigated the biochemical functions of vitamin E and its effects on various bodily systems, highlighting that a deficiency in this lipophilic compound can lead to serious health consequences, while excessive consumption may cause hypervitaminosis. Simultaneously, the researchers pointed to the potent antioxidant effect and pharmacokinetic properties of alpha-tocopherol. Furthermore, they explained the significant influence of the vitamin on the immune, cardiovascular, and nervous systems, on skin health, and its role in the prevention of carcinogenesis.

Concurrently, researchers focused on the effects of B vitamins on the body. C. Munteanu *et al.* (2024) found that vitamin $B_1$ is essential for the body as a cofactor in carbohydrate metabolism, nucleotide synthesis, and the production of nicotinamide adenine dinucleotide. Moreover, the important role of vitamin $B_2$ in maintaining cellular energy balance, growth and development, and metabolism has been established. Vitamin $B_2$ achieves this function as a component of two primary coenzymes: flavin mononucleotide and flavin adenine dinucleotide, which are crucial components that facilitate biochemical intracellular reactions. Thus, based on an analysis of the literature, it can be concluded that the administration of pro- and prebiotics to animals, containing lactic acid bacteria, amino acids, and vitamin preparations, has a positive impact on blood composition, the state of the gut microbiota, strengthens the central nervous system, enhances immunity, and contributes to increased animal productivity.

## Materials and Methods

The experimental studies were conducted throughout 2024 in the research laboratory of immunology of agricultural animals at Bila Tserkva National Agrarian University and the laboratory of anaerobic infections at the Institute of Veterinary Medicine of the National Academy of Agrarian Sciences of Ukraine. The experiment involved 40 white laboratory mice (*Mus musculus* L.), specifically improved conventional animals (*Minimal diseases*) that were free from pathogenic microflora and housed with barrier elements within an improved conventional system. The experiment also included 40 young rabbits of the Grey Giant breed. The laboratory animals (white mice and rabbits) were divided into a control group (20 animals) and an experimental group (20 animals).

In the experiment, white laboratory mice aged between 14 and 60 days were used. The prebiotic Bio-active was administered at a dose of 1.2 g per animal, once daily for 30 days, and the study continued for an additional 30 days after the prebiotic administration ceased. Rabbits were fed the prebiotic at a dose of 1.2 g per animal from 45 to 90 days of age. The diet for the white mice consisted of crushed wheat grain and compound feed, while the rabbits were fed legume hay (80 g), fodder beet (110 g), and compound feed (80 g) per animal. Drinking water for the experimental animals was provided through nipple drinkers.

The investigated prebiotic comprised the metabolic products of the lactic acid bacterium *Lactobacillus bulgaricus delbrueckii*, adsorbed onto zeolite, along with the following amino acids: aspartic acid – 33.77 mg/%, glutamic acid – 10.51 mg/%, glycine – 10.59 mg/%, and phenylalanine – 4.01 mg/%; and the following vitamins: $B_1$ – 0.13 μg/g, $B_2$ – 0.17 μg/g, and $B_{12}$ – 0.0012 μg/g; vitamin A – 0.627 μg/g, and vitamin E – 3.0 μg/g.

During the initial phase of the study, the housing conditions for the white laboratory

mice and rabbits were established. Measurements of microclimate parameters within the vivarium were taken three times throughout the experiment. The vivarium of Bila Tserkva National Agrarian University is situated 1,500 metres from the city of Bila Tserkva. The vivarium grounds are landscaped with greenery. The underground groundwater level was more than 2 metres below the foundation's lowest point. The experimental animals were housed in the vivarium following current regulations (VNTP-APK-05.07..., 2007).

To evaluate the microclimate parameters within the experimental animal housing, the following were determined: temperature (°C), using a TFA 12300802 mercury thermometer (Germany); relative humidity (%), using a VIT-2 psychrometric hygrometer (Ukraine); air velocity (m/s), using a globe katathermometer (Ukraine); the concentration of harmful gases (carbon dioxide, %; ammonia and hydrogen sulphide, mg/m$^3$), using a UG-2 gas analyser (Ukraine); and artificial illumination (lux), using a WT81B lux meter (Wintact, Japan).

The determined microclimate parameters in the vivarium (Table 1), where the white laboratory mice were housed, were within the sanitary and hygienic standards (SOU 85.237736:2011..., 2011). Ventilation was natural – through open doors and a window (or transom). The quality indicators of the water provided to the animals met the requirements of the current State Sanitary Standards – DSanPiN 2.2.4-171-10 (State sanitary norms..., 2010), as evidenced by the data presented in Table 2.

**Table 1**. Microclimate indicators in the vivarium for housing white laboratory mice

| Parameter | Sanitary and hygienic norms | Actual indicator |
|---|---|---|
| Temperature, °C | 20-24 | 22.50 ± 1.48 |
| Relative humidity, % | 55 | 53.80 ± 1.54 |
| Air velocity, m/s | 0.3 | 0.22 ± 0.03 |
| $CO_2$ concentration, % | 0.15 | 0.13 ± 0.01 |
| $NH_3$ concentration, mg/m$^3$ | 10 | 0.08 ± 0.06 |
| $H_2S$ concentration, mg/m$^3$ | 5 | 0.006 ± 0.001 |
| Illumination, lux (1 m from the floor) | 200 | 197.32 ± 6.89 |
| Photoperiod, hours (light: dark) | 12:12 | 12:12 |
| Air exchange rate (ACH) | 10-15 | 12 |

***Source**: authors' development*

**Table 2**. Organoleptic, physicochemical and epidemiological safety indicators of tap water for white laboratory mice

| Parameter | Unit of measurement | DSanPiN 2.2.4-171-10 | Actual indicator |
|---|---|---|---|
| Organoleptic indicators | | | |
| Odour (at t = 20 °C) | points | ⩽ 2 | 1.3 ± 0.1 |
| Turbidity | mg/dm$^3$ | ⩽ 1.0 | 0.4 ± 0.1 |
| Colour | degrees | ⩽ 20.0 | 6.8 ± 0.1 |
| Taste and aftertaste | points | ⩽ 2 | 1.4 ± 0.4 |
| Physicochemical indicators | | | |
| Hydrogen index | pH | 6.5–8.5 | 6.9 ± 0.1 |
| Total hardness | mmol/dm$^3$ | ⩽ 7.0 | 6.8 ± 0.2 |

Table 2. Continued

| Parameter | Unit of measurement | DSanPiN 2.2.4-171-10 | Actual indicator |
|---|---|---|---|
| Physicochemical indicators | | | |
| Chlorides | mg/dm³ | ⩽ 250 | 196.0 ± 5.6 |
| Nitrates | mg/dm³ | ⩽ 50 | 15.0 ± 3.2 |
| Nitrites | mg/dm³ | ⩽ 0.5 | 0.4 ± 0.02 |
| Manganese | mg/dm³ | 0.1 | 0.03 ± 0.01 |
| Sulphates | mg/dm³ | ⩽ 250 | 38.4 ± 2.5 |
| Total iron | mg/dm³ | ⩽ 0.2 | 0.08 ± 0.003 |
| Epidemiological safety indicators | | | |
| Total coliforms | CFU/100 cm³ | absent | absent |
| Intestinal helminths | Cells, eggs, larvae in 50 dm³ | absent | absent |

***Source**: authors' development*

The vivarium where the rabbits were housed was a standard, single-storey building constructed from white brick, located on level ground with good natural sunlight. The control and experimental groups of rabbits were kept in four wire mesh cages (10 animals in each). Each cage was equipped with hay racks, trough feeders, and a trough drinker. The examination of individual physical parameters of the microclimate in the vivarium (Table 3), where the rabbits were housed, indicated that they met the sanitary and hygienic requirements of the document – Departmental Standards for Technological Design of Enterprises of Animal Husbandry and Rabbit Breeding (VNTP-APK-05.07, 2008). Lighting in the room was both natural and artificial – fluorescent lamps were used. Ventilation in the animal housing was natural – through open doors and a window (or transom). The measured indicators of the water (Table 4) provided to the control and experimental groups of rabbits throughout the experiment met the requirements of the current State Sanitary Standards – DSanPiN 2.2.4-171-10 (State sanitary norms..., 2010).

**Table 3.** Microclimate parameters in the vivarium for housing rabbits

| Parameter | Sanitary and hygienic norms | Actual indicator |
|---|---|---|
| Temperature, °C | 15-20 | 20.0 ± 1.15 |
| Relative humidity, % | 55 | 52.4 ± 1.36 |
| Air velocity, m/s | 0.3 | 0.26 ± 0.01 |
| $CO_2$ concentration, % | 0.15 | 0.10 ± 0.03 |
| $NH_3$ concentration, mg/m³ | 10 | 0.82 ± 0.02 |
| $H_2S$ concentration, mg/m³ | 5 | 0.007 ± 0.001 |
| Illumination, lux (1 m from the floor) | 200 | 187.26 ± 2.57 |
| Photoperiod, hours (light: dark) | 12:12 | 12:12 |
| Air exchange rate (ACH) | 10-15 | 10-15 |

***Source**: authors' development*

**Table 4**. Organoleptic, physicochemical, and epidemiological safety indicators of tap water for rabbits

| Parameter | Unit of measurement | DSanPiN 2.2.4-171-10 | Actual indicator |
|---|---|---|---|
| Organoleptic indicators | | | |
| Odour (at t = 20 °C) | points | ⩽ 2 | 1.6 ± 0.1 |
| Turbidity | mg/dm$^3$ | ⩽ 1.0 | 0.2 ± 0.04 |
| Colour | degrees | ⩽ 20.0 | 5.3 ± 0.1 |
| Taste and aftertaste | points | ⩽ 2 | 1.6 ± 0.2 |
| Physicochemical indicators | | | |
| Hydrogen index | pH | 6.5–8.5 | 7.2 ± 0.3 |
| Total hardness | mmol/dm$^3$ | ⩽ 7.0 | 5.9 ± 0.8 |
| Chlorides | mg/dm$^3$ | ⩽ 250 | 200.3 ±7.9 |
| Nitrates | mg/dm$^3$ | ⩽ 50 | 13.6 ± 6.9 |
| Nitrites | mg/dm$^3$ | ⩽ 0.5 | 0.2 ± 0.1 |
| Manganese | mg/dm$^3$ | 0.1 | 0.02 ± 0.01 |
| Sulphates | mg/dm$^3$ | ⩽ 250 | 42.3 ± 5.2 |
| Total iron | mg/dm$^3$ | ⩽ 0.2 | 0.07 ± 0.01 |
| Epidemiological safety indicators | | | |
| Total coliforms | CFU/100 cm$^3$ | absent | absent |
| Intestinal helminths | Cells, eggs, larvae in 50 dm$^3$ | absent | absent |

***Source**: authors' development*

During the second stage of the study, the impact of the prebiotic on the dynamics of growth and development, as well as the survival rate of white laboratory mice and rabbits, was assessed. Body weight gain in the laboratory mice and rabbits following prebiotic administration was determined by measuring body weight at the start and end of the study, and by calculating the average daily weight gain using standard calculation methods. To further investigate the effects of the studied prebiotic on the white mice, the mass of internal organs – thymus, kidneys, thyroid gland, liver, and spleen – was determined using standard procedures.

The third stage of the study examined the effect of the prebiotic on haematological parameters in white laboratory mice and rabbits. For the haematological analyses, peripheral blood from white laboratory mice was collected by decapitation on the 60$^{th}$ day of the experiment. Prior to this, the white mice were anaesthetised with 2% xylazine (Netherlands) and then decapitated using a guillotine. In rabbits, blood samples were collected from the marginal ear vein on the 90$^{th}$ day of the study, before the morning feed.

A full blood count of the experimental animals was conducted using an HTI MicroCC-25 Plus automated haematology analyser (USA). The parameters determined were: red blood cell count (RBC, $10^{12}$/L), haemoglobin concentration (HGB, g/L), haematocrit (HCT), white blood cell count (WBC, $10^9$/L), neutrophils (NEUT, %), lymphocytes (LYMPH, %), basophils (BASO, %), eosinophils (EO, %), and monocytes (MONO, %). Biochemical parameters of the blood serum of the experimental animals were determined using an AS-120 automated biochemical analyser (Japan) and a test system from Global Scientific (USA).

During the fourth stage of the study, the effect of the prebiotic Bio-active on the quantitative composition of the intestinal microflora in rabbits was analysed. Rabbits were culled

at 90 days of age (DSTU 4293:2004, 2004), and smears were prepared from the caecal mucosa onto microscope slides and stained using the Gram method. Subsequently, the number of Gram-negative and Gram-positive microorganisms was counted (Laukens *et al.*, 2016).

The studies involving laboratory animals were conducted following the requirements of the General Ethical Principles of Animal Experiments adopted by the First National Congress on Bioethics (Kyiv, 2001) (On the protection…, 2021) and in compliance with the international principles outlined in the European Convention for the Protection of Vertebrate Animals used for Experimental and other Scientific Purposes (European Convention, 1986) and the Law of Ukraine "On the Protection of Animals from Cruelty" (Law of Ukraine No. 3447-IV, 2006).

All obtained numerical data were statistically processed using the Microsoft Excel software package. The following were calculated: the arithmetic mean and its error (M ± m), the level of statistical significance (P) using Student's t-test, and the significance criteria were defined as: $P < 0.05$; $P < 0.01$; $P < 0.001$.

## Results and Discussion

Research into the effects of preparations, particularly the prebiotic Bio-active (which contains a range of components aimed at maintaining natural resistance, growth energy, blood condition, and gut microbiome), on the bodies of laboratory animals should commence with an analysis of body weight and internal organ dynamics. The body weight trends of the experimental animals can indicate whether the condition of their organs is within the physiological norm. The study of body weight and internal organ dynamics in white laboratory mice is presented in Table 5.

**Table 5**. Body weight and internal organ mass in white laboratory mice after prebiotic feeding, M ± m, n = 20

| Parameter, unit of measurement | The day of the study | | | |
|---|---|---|---|---|
| | 1 | 14 | 30 | 60 |
| Animal body weight, g | 11.0 ± 0.85<br>11.0 ± 0.64 | 14.0 ± 0.74<br>15.8 ± 0.85▲ | 17.0 ± 0.94<br>20.0 ± 0.79*▲ | 22.1 ± 1.03<br>25.6 ± 1.08*▲ |
| Average daily weight gain, mg | – | 214.12 ± 1.06<br>341.10 ± 0.85** | 186.52 ± 1.72<br>260.45 ± 2.01** | 167.86 ± 1.86<br>188.56 ± 1.90** |
| Thymus mass, mg | – | | – | 30.0 ± 3.0<br>34.0 ± 2.3 |
| Liver mass, g | – | – | – | 1.14 ± 0.04<br>1.18 ± 0.05 |
| Spleen mass, g | – | – | – | 0.19 ± 0.03<br>0.19 ± 0.02 |
| Thyroid gland mass, mg | – | – | – | 4.0 ± 0.05<br>4.0 ± 0.06 |
| Kidney mass, g | – | – | – | 0.14 ± 0.07<br>0.14 ± 0.03 |

***Note:*** *numerator – control group; denominator – experimental group; *P<0.05; **P<0.001, compared to the control group. ▲P<0.001, compared to the indicator at the beginning of the study; 1 – start of the experiment*
***Source****: authors' development*

According to the results presented in Table 5, by day 14, the body weight of the white mice had increased by 43.6% ($P < 0.001$) compared to the body weight of the mice at the start of the study. Furthermore, the average daily weight gain in the mice had increased by 59.3% ($P < 0.001$) compared to the animals in the control group. By day 30 of the study, the body weight of the white mice had increased by 17.65% ($P < 0.05$) compared to the control

group, and by 81.82% (P < 0.001) compared to the body weight at the start of the study. The average daily weight gain in the mice had increased by 39.64% (P < 0.001) compared to the control group. By day 60 of the study, the body weight in the white mice had increased by 15.84% (P < 0.05) compared to the control group, and by 132.73% (P < 0.001) compared to this indicator at the start of the study. The average daily weight gain in the white mice had increased by 12.33% (P < 0.001) compared to the control group. Throughout the entire study period, the animals actively consumed both the feed from the basic diet and the feed supplemented with the prebiotic. The behaviour of the experimental animals was normal – they were mobile and active. Table 5 indicates that there was no statistically significant difference in the mass of the studied internal organs of the laboratory mice between the experimental and control groups.

As the studied prebiotic is a complex preparation containing live cultures of lactic acid bacteria, amino acids, and vitamins A, E, and B complex, its effect on the animals' bodies depends on the symbiotic influence of these components. Specifically, C. Li *et al.* (2024) demonstrated that synbiotics regulate metabolism, reduce insulin resistance, and positively influence body weight dynamics in mice. The current study established that the prebiotic Bio-active also promoted growth and development in Grey Giant rabbits, with a 100% survival rate (Table 6).

**Table 6**. Growth parameters in experimental rabbits, M ± m, n = 20

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| Number of animals, heads: | | |
| ♦ at the start of the study | 20 | 20 |
| ♦ at the end of the study | 20 | 20 |
| Survival rate, % | 100 | 100 |
| Live weight of animals, kg: | | |
| ♦ at the start of the study | 0.965 ± 0.068 | 0.983 ± 0.057 |
| ♦ at the end of the study | 2.185 ± 0.024 | 2.461 ± 0.024 |
| Absolute weight gain of the entire group, kg | 1.220 | 1.478 |
| Relative weight gain per animal, % | 126.4 | 150.3 |
| Growth energy, g | 40.6 ± 1.93 | 49.2 ± 1.64* |
| Morbidity, % | – | – |
| Condition of the coat | Normal | Normal |

***Note:*** **P < 0.01, compared to the control group*
***Source****: authors' development*

According to the body weight measurements taken at the beginning and end of the study, no statistically significant difference was found between the groups. However, the absolute weight gain in all rabbits in the experimental group was 21.15% higher than that of the control group, and the relative weight gain per rabbit in the experimental group was 23.9% higher compared to the control group. It was found that the growth energy of rabbits in the experimental group increased by 21.2% (P < 0.01) compared to the animals in the control group. Thus, the daily feeding of the prebiotic Bio-active at a dose of 1.2 g per animal for 45 days contributed to improved survival, metabolism, and accelerated growth, energy and development in Grey Giant rabbits.

The positive outcomes observed with the use of the prebiotic in rabbits were dependent on the meticulous control of both the

preparation dosage and the feeding ration. In this regard, Y. Shastak & W. Pelletier (2023) demonstrated that research should focus on determining the optimal periods for adjusting feeding rations and the dosage of supplements. Furthermore, considering that the prebiotic contains biologically active components, including vitamin A, scientists highlighted its recognised properties as a potent antioxidant, essential for animals to maintain their productivity levels and immunity.

One of the key stages of the study was the evaluation of the blood condition of the experimental animals. After all, the blood parameters of animals can reveal the functional state of the body, allow for the timely detection of signs of inflammation, and also confirm that the control group of animals consisted of healthy white mice (which naturally maintain a relatively constant blood composition) and rabbits. Therefore, it is very important to determine the dynamics of blood parameters in both white laboratory mice and rabbits during the feeding of the studied prebiotic. The results of its effect on the morphological parameters and leukogram of the blood of white mice are presented in Table 7.

**Table 7**. Complete blood count in white laboratory mice, M ± m, n = 20

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| Red blood cells (RBC), $10^{12}$/L | 7.98 ± 0.15 | 8.49 ± 0.12* |
| Haemoglobin (HGB), g/L | 89.0 ± 3.16 | 98.0 ± 3.02* |
| White blood cells (WBC), $10^{9}$/L | 7.60 ± 0.40 | 7.50 ± 0.50 |
| Band neutrophils (NEUT), % | 0.54 ± 0.21 | 0.78 ± 0.36 |
| Segmented neutrophils (NEUT), % | 21.76 ± 1.65 | 19.43 ± 2.86 |
| Basophils (BASO), % | 0.30 ± 0.01 | 0.28 ± 0.02 |
| Lymphocytes (LYMPH), % | 55.0 ± 4.12 | 57.43 ± 3.02 |
| Eosinophils (EO), % | 2.98 ± 0.24 | 2.45 ± 0.16 |
| Monocytes (MONO), % | 5.90 ± 2.45 | 5.60 ± 1.02 |

***Note:*** **P < 0.05, compared to the control group*
***Source***: *authors' development*

Table 7 shows that in the blood of white laboratory mice, the red blood cell count increased by 6.4% (P < 0.05), and the haemoglobin concentration by 10.1% (P < 0.05) compared to the control group. As the obtained research results indicate, the preparation activates erythropoiesis processes in the bodies of white laboratory mice. Furthermore, its administration for 30 days, at a dose of 1.2 g per animal, does not suppress the myeloid and lymphoid lineages of blood cell formation. The effect of the preparation continues up to 60 days of age in the laboratory mice. The biochemical parameters of the blood serum in white laboratory mice are presented in Table 8.

**Table 8**. Biochemical indicators of blood serum in white laboratory mice, M ± m, n = 20

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| Total protein (T-Pro), g/L | 63.0 ± 2.20 | 70.0 ± 2.06* |
| Albumins (ALB), % | 55.0 ± 3.18 | 57.0 ± 2.10 |
| Globulins (Glob), % | 8.10 ± 0.64 | 11.0 ± 0.82** |
| Alanine aminotransferase (ALT), U/L | 45.38 ± 2.46 | 46.23 ± 2.67 |

Table 8. Continued

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| Aspartate aminotransferase (AST), U/L | 124.61 ± 5.32 | 127.08 ± 6.89 |
| Total cholesterol, mmol/L | 2.14 ± 0.3 | 2.12 ± 0.2 |

***Note:*** **P < 0.05; **P < 0.01, compared to the control group*
***Source****: authors' development*

The research results showed that under the influence of the prebiotic, the total protein content in the blood serum of white mice increased by 11.1% (P < 0.05) and globulins by 1.4 times (P < 0.01) compared to the control group. It can be suggested that the increase in total protein and globulin levels in the blood serum of the laboratory mice is are key factor in enhancing the growth energy and development of the animals. Thus, the obtained research results indicate an increase in the activity of the humoral immunity component in the experimental group of animals.

The increase in total protein content in the blood serum of white laboratory mice can also be attributed to the additional intake of proteinogenic amino acids present in the studied preparation. In particular, Y. Kamei *et al.* (2020) found that amino acids, acting as biological regulators, stimulate the anabolism of muscle proteins and enhance resistance in animals. Furthermore, no statistically significant difference was found between the activity levels of aspartate aminotransferase and alanine aminotransferase in the blood serum of mice in the experimental and control groups.

Based on the results of the cholesterol content study in the blood serum of the laboratory mice, it can be noted that the changes in metabolism in the experimental group animals occurred due to anabolic processes, as no statistically significant difference was observed for this parameter between the experimental and control groups of mice. However, S. Ghosh *et al.* (2020) demonstrated that the addition of prebiotics to the diet of mice led to a decrease in serum cholesterol levels, irrespective of the animals' body weight changes.

Determining the blood condition of rabbits in the experiment is crucial for a timely response to changes in the functioning of the animals' bodies. Specifically for rabbits, the blood condition can indicate the presence of bacterial, viral, fungal, or parasitic infections, disturbances in the functioning of the gastrointestinal tract, and consequently, a weakening of the immune system. The effect of the prebiotic on the morphological and biochemical parameters of rabbit blood is presented in Table 9.

**Table 9**. Haematological indicators in experimental rabbits, M ± m, n = 20

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| Red blood cells (RBC), $10^{12}$/L | 4.85 ± 0.14 | 5.42 ± 0.16* |
| Haemoglobin (HGB), g/L | 112.43 ± 4.12 | 125.32 ± 3.56* |
| ESR, mm/hour | 4.12 ± 0.50 | 3.90 ± 0.30 |
| White blood cells (WBC), $10^{9}$/L | 6.80 ± 0.40 | 6.56 ± 0.10 |
| Total protein (T-Pro), g/L | 59.0 ± 1.15 | 65.0 ± 2.16* |
| Albumins (ALB), % | 29.40 ± 2.12 | 30.0 ± 1.84 |
| Globulins (Glob), % | 29.6 ± 1.12 | 35.0 ± 1.91* |

Table 9. Continued

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| Alanine aminotransferase (ALT), U/L | 8.20 ± 0.21 | 9.80 ± 0.14 |
| Aspartate aminotransferase (AST), U/L | 24.0 ± 0.98 | 28.0 ± 0.64 |
| Total cholesterol, mmol/L | 1.96 ± 0.03 | 1.90 ± 0.02 |

***Note:*** **P < 0.05, compared to the control group*
***Source****: authors' development*

The results of the experimental studies (Table 9) established that after feeding the rabbits with the prebiotic, activation of the erythropoiesis process was observed, as evidenced by an increase in the number of red blood cells in the blood by 11.7% P < 0.05), an increase in haemoglobin concentration by 11.5% (P < 0.05), total protein by 10.2% (P < 0.05), and globulins by 1.2 times (P < 0.05) compared to the control group. It is worth noting that this may be the main factor in increasing the growth energy and development of rabbits. No statistically significant difference was found for the remaining parameters. All investigated parameters were within the reference values.

N.B. Abduljabbar *et al.* (2024) found that adding a prebiotic – algae powder – to the diet of rabbits increased (P < 0.05) in body weight, blood haemoglobin content, red blood cell count, total protein concentration, albumin, and high-density lipoproteins, alongside a decrease (P < 0.05) in white blood cell count and cholesterol concentration. Similar results were obtained by A. Mohammed (2023), who found that adding enzymes to rabbit feed increased the red blood cell count (P < 0.05) and haemoglobin content. In turn, M. Sobri *et al.* (2019) demonstrated that the particle size of fibre in a feed supplement had a significant effect (P < 0.05) on the number of white blood cells, eosinophils, and neutrophils in the blood of post-weaning rabbits. The data from these mentioned studies align with the results of the current experiment. The increase, within the physiological range, of enzymatic activity indicators after feeding the studied prebiotic suggested the absence of liver damage and necrosis, with a moderate stimulation of metabolism.

According to the results of the study on the effect of the prebiotic on the concentration of total cholesterol in the blood serum of rabbits, it was noted that the changes in metabolism in the experimental animals occurred due to anabolic processes, as no statistically significant difference was observed for this parameter between the experimental and control groups of animals. Therefore, the prebiotic contributed to the activation of the energy and structural needs of the experimental group rabbits and stimulated the main metabolic pathways of their functioning.

The studied prebiotic contains the lactic acid bacterium *Lactobacillus bulgaricus delbrueckii*, which inhibits the proliferation of opportunistic and pathogenic microflora and can create conditions for lowering the pH in the animals' intestines. Therefore, this paper presents the results of an evaluation of the effect of the lactic acid bacterium *Lactobacillus bulgaricus delbrueckii*, in combination with other bioactive components of the Bio-active prebiotic, on the percentage composition of the microflora in the caecum of experimental rabbits. According to the results of the performed microscopy of smears (Table 10) prepared from the caecal mucosa of rabbits in the experimental group, it was found that the total number of Gram-positive microorganisms increased by 19.4% (P < 0.05), while Gram-negative microorganisms, conversely, decreased by 17.7% (P < 0.05) compared to the control group.

**Table 10**. Microscopy of smears from the caecal mucosa of rabbits, M ± m, n = 10

| Parameter, unit of measurement | Control group | Experimental group |
|---|---|---|
| pH value | 7.1 ± 0.2 | 6.5 ± 0.2 |
| Gram-positive microflora, % | 48.0 ± 5.0 | 67.4 ± 2.6* |
| Gram-negative microflora, % | 39.0 ± 4.6 | 21.3 ± 1.3* |
| Ratio of Gram-negative to Gram-positive | 1:1.23 | 1:3.16 |

***Note:*** **P < 0.05, compared to the control group*
***Source****: authors' development*

Furthermore, the Gram-positive microorganisms were observed as thick, long rods, while the Gram-negative ones were small, thin, and long rods with granularity. Coccal forms of Grampositive bacteria (streptococci, staphylococci, and micrococci) were also observed in the microscope's field of view. In the caecal microbiome of rabbits in the experimental group, the proportion of Gram-positive microflora predominated over Gram-negative microflora, which is a physiological phenomenon for the large intestine, ensuring a balanced structural and energy metabolism.

Table 10 shows that there was no statistically significant difference in the pH value of the caecal contents of rabbits between the experimental and control groups. However, in the rabbits of the experimental group, the pH of the caecal contents was 6.5 (slightly acidic). In contrast, in the rabbits of the control group, the pH value corresponded to a neutral reaction (pH 7.1). In this regard, S.H. Abu Hafsa *et al.* (2022) demonstrated that the pH value of the rabbit caecum is within a range that ensures the activity of digestive enzymes in the large intestine and improves digestion and nutrient absorption. The authors found that the pH value of the caecal mucosa ranged from 6.0 to 6.6. If the pH value is too high (greater than 7.5), it will affect the activity of digestive enzymes and lead to the proliferation of pathogenic bacteria, which may provoke the onset of diarrhoea in rabbits. The appropriate pH range of the intestine is maintained, as noted by A.B. Negussie *et al.* (2022), by the epithelial cells of the large intestine. These cells establish osmotic gradients, which are key for retaining fluid and maintaining the level of secretion, as well as regulating cell death. Furthermore, S. Li *et al.* (2023) found that the ratio of fibre content in the diet of rabbits affects the pH value in the caecum. This influences the immunomodulatory function of the intestine and the level of nutrient absorption.

During the experiment, it is important to adhere not only to a specific dose of the preparation but also to the age periods of the experimental animals, which are associated with the transition to a different type of feed. Therefore, to investigate the quantity of cecal microbiota, rabbits aged 45 days were used, that is, during the weaning period and the transition to a plant-based diet. S.M. Leite *et al.* (2024) emphasised that after weaning rabbit kits (from 40 days of age) from the mother, a series of physiological changes occur in the gastrointestinal tract depending on the type of feed. T. Read *et al.* (2023) also indicated that from the 43$^{rd}$ day of life in rabbits consuming solid feed, the structure and interrelationships within the gut microbiome change. Furthermore, E. Cotozzolo *et al.* (2021) demonstrated that the greatest diversity of the microbiome is found in the caecum and throughout the large intestine of rabbits. In particular, clarifying the quantity and species-specific characteristics of the rabbit gut microbiota will help improve their health and contribute to increased productivity.

The establishment of the complex effect of the components of the studied prebiotic on the ratio of microflora in the rabbit caecum proved to be positive. It can be suggested that due to the presence of the lactic acid bacterium strain *Lactobacillus bulgaricus delbrueckii* in the studied prebiotic, such a ratio of microflora in the large intestine was maintained, which contributed to the preservation of growth, development, and overall health indicators in rabbits. K. Song *et al.* (2024) noted that one of the representatives of the beneficial gut microflora involved in the restoration of microflora is the lactic acid bacterium strain – *Lactobacillus delbrueckii* subsp. *Bulgaricus*. The authors demonstrated the beneficial effect of this strain on the experimentally damaged mucosa of the large intestine in mice, as well as the restoration of the intestinal microbiota and the improvement of the intestinal condition. C. Zhao *et al.* (2021) found that supplementing the feed of lactating rabbits with *B. subtilis*, *B. Licheniformis*, and *S. cerevisiae* increases the populations of beneficial gut bacteria, improves nutrient digestibility, enhances fermentation in the caecum, increases feed conversion, and promotes body weight gain. Thus, supplements with live bacterial cultures in rabbit feed are an alternative to antibiotics and significantly affect the improvement of productivity, growth, prevention and treatment of intestinal diseases, as well as increasing animal immunity.

Overall, the positive effect of the studied prebiotic on the bodies of white laboratory mice and rabbits was manifested as a successful symbiotic action of the lactic acid bacterium *Lactobacillus bulgaricus delbrueckii* together with amino acids and vitamins. J.E. Nettleton *et al.* (2021) demonstrated that the prevention and treatment of mice with prebiotics, probiotics, and synbiotics altered the species composition of their gut microbiota and led to improved intestinal function. In this context, A. Bevilacqua *et al.* (2024) presented the updated definition of prebiotics by a consensus panel of the International Scientific Association for Probiotics and Prebiotics (ISAPP): “prebiotics are substrates that are selectively utilized by host microorganisms conferring a health benefit”.

Thus, the complex prebiotic, when fed to white laboratory mice and Grey Giant rabbits, contributed to an increase in body weight, red blood cell count, haemoglobin and globulin levels, and liver enzyme activity, as well as a decrease in cholesterol levels. Furthermore, the percentage composition of Gram-positive microflora in the caecum of rabbits predominated over Gramnegative microflora. The validity of the experimental studies was confirmed by the indicators of the microclimate and the drinking water provided to the laboratory animals (white mice and rabbits), which met the requirements of the current regulatory documents. The presented research results confirm the need to consider the age of the experimental animals, the type of their feed, and the dosage of the prebiotic. This scientific approach is important for the further practical application of the obtained results, for maintaining animal health, and for reducing mortality rates.

## Conclusions

The conducted study has revealed the positive effect of the components of the Bio-active prebiotic at a dose of 1.2 g per animal on the bodies of white laboratory mice and rabbits. It was established that the microclimate parameters in the vivarium where the white mice and rabbits were kept, as well as the indicators of drinking water provided to them, met the requirements of the current regulatory documents. Feeding the Bio-active prebiotic at a dose of 1.2 g per animal contributed to an increase in the body weight of white mice in the experimental group compared to the control group throughout the

entire study period (day 14 by 43.6% ($P < 0.001$); day 30 by 17.65% ($P < 0.05$); day 60 by 15.84% ($P < 0.05$)), as well as compared to the indicators at the beginning of the study (day 14 by 43.6% ($P < 0.001$); day 30 by 81.82% ($P < 0.001$); day 60 by 132.73% ($P < 0.001$)). During the experiment, the average daily weight gain of white mice in the experimental group also increased (day 14 by 59.3% ($P < 0.001$); day 30 by 39.64% ($P < 0.001$); day 60 by 12.33% ($P < 0.001$)). The mass of the studied internal organs (thymus, liver, spleen, thyroid gland, and kidneys) in the laboratory mice did not change significantly between the experimental and control groups of animals. The studied prebiotic at a dose of 1.2 g per animal positively influenced the increase in body weight, absolute and relative weight gain in rabbits, which improved survival rates and growth energy. The prebiotic contributed to the activation of erythropoiesis in the bodies of the experimental white mice and rabbits. In the blood of white mice, the red blood cell count and haemoglobin concentration increased by 6.4% ($P < 0.05$) and 10.1% ($P < 0.05$), respectively, and in the blood of rabbits – by 11.7% ($P < 0.05$) and 12.6% ($P < 0.05$), respectively. The effect of the prebiotic on the bodies of the experimental animals was expressed in an increased activity of the humoral immunity component, which manifested in an increase in the blood serum of white mice in the content of total protein by 11.1% ($P < 0.05$) and globulins by 1.4 times ($P < 0.01$), and in rabbits – by 10.2% ($P < 0.05$) and 1.2 times ($P < 0.05$), respectively. The activity of aspartate aminotransferase and alanine aminotransferase, and the cholesterol content in the blood serum of white mice and rabbits in the experimental and control groups, remained unchanged. A positive effect of the studied prebiotic on the composition of the caecal microbiome of rabbits in the experimental group was established, which contributed to an increase in the total number of Gram-positive microorganisms by 19.4% ($P < 0.05$) and a decrease in Gram-negative ones by 17.7% ($P < 0.05$). The effect of the preparation at a dose of 1.2 g per animal on the bodies of white laboratory mice and rabbits contributed to an increase in the indicators of growth and development of the body, and the functional capacity of the large intestine, which affects the maintenance of animal health.

The obtained results of the experimental study may be useful for clarifying the complex effect of the supplement components on the bodies of laboratory animals. In the future, further research will be devoted to determining the species composition of the large intestine microbiome in rabbits.

## Acknowledgements

None.

## Funding

The study received no funding.

## Conflict of Interest

None.

## References

[1] Abduljabbar, N.B., Aljanabi, A.A.F., & Alslami, M.S.M. (2024). Effect of different levels on some physiological and productive parameters of local rabbits in Iraq. *IOP Science*, 1371, article number 072034. doi: 10.1088/1755-1315/1371/7/072034.

[2] Abu Hafsa, S.H., Mahmoud, A.E.M., Fayed, A.M.A., & Abdel-Azeem, A.-A.S. (2022). The effect of exogenous lysozyme supplementation on growth performance, caecal fermentation and microbiota, and blood constituents in growing rabbits. *Animals*, 2(7), article number 899. doi: 10.3390/ani12070899.

[3] Asadi, M, Fard, H.M., Araee, K.A., & Hatami, M. (2024). Studying the impacts of maternal B complex vitamin injection on performance, metabolic diseases, hematological parameters, and antioxidant status in pregnant Sannen goats and their newborn kids during the transition period. *Science of the Total Environment*, 907, article number 167860. doi: 10.1016/j.scitotenv.2023.167860.

[4] Ballini, A., Charitos, I.A., Cantore, S., Topi, S., Bottalico, L., & Santacroce, L. (2023). About functional foods: The probiotics and prebiotics state of art. *Antibiotics*, 12(4), article number 635. doi: 10.3390/antibiotics12040635.

[5] Beaumont, M., Roura, E., Lambert, W., Turni, C., Michiels, J., & Chalvon-Demersay, T. (2022). Selective nourishing of gut microbiota with amino acids: A novel prebiotic approach? *Frontiers in Nutrition*, 9, article number 1066898. doi: 10.3389/fnut.2022.1066898.

[6] Bevilacqua, A., Campaniello, D., Speranza, B., Racioppo, A., Sinigaglia, M., & Corbo, M.R. (2024). An update on prebiotics and on their health effects. *Foods*, 13(3), article number 446. doi: 10.3390/foods13030446.

[7] Biben, I.A., Sosnitskyi, O.I., & Zazharskyi, V.V. (2021). Subaerin symbiotic: Composition, biological effect on macroorganism. *Scientific and Technical Bulletin* of *State Scientific Research Control Institute of Veterinary Medical Products and Fodder Additives* and *Institute of Animal Biology*, 22(2), 55-65. doi: 10.36359/scivp.2021-22-2.06.

[8] Burmei, S.A., & Boyko, N.B. (2024). Key aspects of the use of new-generation probiotic strains in 3P medicine: Challenges and perspectives. *Intermedical Journal*, 1, 28-36. doi: 10.32782/2786-7684/2024-1-5.

[9] Cotozzolo, E., *et al.* (2021). Characterization of bacterial microbiota composition along the gastrointestinal tract in rabbits. *Animals*, 11(1), article number 31. doi: 10.3390/ani11010031.

[10] De Jesus, L.L.C., *et al*. (2024). Health-promoting effects and safety aspects of *Lactobacillus delbrueckii*: A food industry species. *Trends in Food Science & Technology*, 150, article number 104605. doi: 10.1016/j.tifs.2024.104605.

[11] DSTU 4293:2004. (2004). *Rabbits for slaughter. Technical conditions*. Retrieved from https://online.budstandart.com/ua/catalog/doc-page?id_doc=74262.

[12] El-Sabrout, K., Khalifah, A., & Ciani, F. (2023). Current applications and trends in rabbit nutraceuticals. *Agriculture*, 13(7), article number 1424. doi: 10.3390/agriculture13071424.

[13] European Convention for the Protection of Vertebrate Animals Used for Experimental and Scientific Purposes. (1986, March). Retrieved from http://zakon4.rada.gov.ua/laws/show/994_137.

[14] Ghosh, S., Yang, X., Wang, L., Zhang, C., & Zhao, L. (2020). Active phase prebiotic feeding alters gut microbiota, induces weight-independent alleviation of hepatic steatosis and serum cholesterol in high-fat diet-fed mice. *Computational and Structural Biotechnology Journal*, 9, 448-458. doi: 10.1016/j.csbj.2020.12.011.

[15] Guzmán-Escalera, D., Valdés-Miramontes, E.H., Iñiguez-Muñoz, L.E., Reyes-Castillo, Z., & Espinoza-Gallardo, A.C. (2025). Metabolites generated from foods through lactic fermentation and their benefits on the intestinal microbiota and health. *Journal of Medicinal Food*, 8(1), 1-11. doi: 10.1089/jmf.2023.0218.

[16] He, W., Song, H., Yang, Z., Zhao, S., Min, J., & Jiang, Y. (2024). Beneficial effect of *GABA*-rich fermented milk whey on nervous system and intestinal microenvironment of aging mice induced by *D*-galactose. *Microbiological Research*, 278, article number 127547. doi: 10.1016/j.micres.2023.127547.

[17] Ishii, I., & Bhatia, M. (2023). Amino acids in health and disease: The good, the bad, and the ugly. *International Journal of Molecular Sciences*, 24(5), article number 4931. doi: 10.3390/ijms24054931.

[18] Jing, J., Jin, W., Liu, S-J., Jiao, Z., & Li, X. (2023). Probiotics, prebiotics, and postbiotics inhealth and disease. *MedComm*, 4, article number e420. doi: 10.1002/mco2.420.

[19] Kamei, Y., Hatazawa, Y., Uchitomi, R., Yoshimura, R., & Miura, S. (2020). Regulation of skeletal muscle function by amino acids. *Nutrients*, 12(1), article number 261. doi: 10.3390/nu12010261.

[20] Kobayashi, K., Mochizuki, J., Yamazaki, F., & Sashihara, T. (2023). Yogurt starter strains ameliorate intestinal barrier dysfunction via activating AMPK in Caco-2 cells. *Tissue Barriers*, 12(1), article number 2184157. doi: 10.1080/21688370.2023.2184157.

[21] Koprivica, M., & Miljković, A. (2024). Impact of vitamin E on different organ systems. *Sanamed*, 19(2), 215-219. doi: 10.5937/sanamed0-49398.

[22] Kunovsky, V.V., Laba, O.V., & Kunovska, L.M. (2024). The role of probiotic cultures in the formation of the intestinal microbiota. The microbiota-gut-brain axis. *Infectious Diseases*, 4, 43-50. doi: 10.11603/1681-2727.2024.4.15006.

[23] Laukens, D., Brinkman, B.M., Raes, J., De Vos, M., & Vandenabeele, P. (2016). Heterogeneity of the gut microbiome in mice: Guidelines for optimizing experimental design. *FEMS Microbiology Reviews*, 40(1), 117-132. doi: 10.1093/femsre/fuv036.

[24] Law of Ukraine No. 3447-IV "On the Protection of Animals from Cruelty". (2006, February). Retrieved from https://zakon.rada.gov.ua/laws/show/3447-15#Text.

[25] Leite, S.M., Santos, E.M G.S., Almeida, M.R., Oliva, N., Stevanato, G.G., Gasque, J.P.N., Ribeiro, L.B., & Castilha, L.D. (2024). Digestive physiology of rabbits in the pre- and post-weaning phases. *Acta Scientiarum. Animal Sciences*, 46(1), article number e70031. doi: 10.4025/actascianimsci.v46i1.70031.

[26] Li, C., Li, D., Liu, H., Zhang, N., Dang, L., Wang, M., Tian, H., Jha, R., & Li, C. (2024). Synbiotic supplementation with xylooligosaccharide derived probiotic *Lactobacillus gasseri* and prebiotic mixture exerts antidiabetic effects via collaborative action. *Food Bioscience*, 61, article number 104948. doi: 10.1016/j.fbio.2024.104948.

[27] Li, S., Liu, T., Wang, K., Li, C., Wu, F., Yang, X., Zhao, M., Chen, B., & Chen, X. (2023). The ratios of dietary non-fibrous carbohydrate (NFC) to neutral detergent fiber (NDF) influence intestinal immunity of rabbits by regulating gut microbiota composition and metabolites. *Frontiers in Microbiology*, 14, article number 1146787. doi: 10.3389/fmicb.2023.1146787.

[28] Mohammed, A. (2023). Effect of multi-enzyme supplementation on growth performance of rabbits. *Asia Pacific Journal of Sustainable Agriculture*, *Food and Energy*, 11(1), 11-15. doi: 10.36782/apjsafe.v11i1.162.

[29] Munteanu, C., Mârza, S.M., & Papuc, I. (2024). The immunomodulatory effects of vitamins in cancer. *Frontiers in Immunology*, 15, article number 1464329. doi: 10.3389/fimmu.2024.1464329.

[30] Negussie, A.B., Dell, A.C., Davis, B.A., & Geibel, J.P. (2022). Colonic fluid and electrolyte transport 2022: An update. *Cells*, 11(10), article number 1712. doi: 10.3390/cells11101712.

[31] Nettleton, J.E., Klancic, T., Schick, A., Choo, A.C., Cheng, N., Shearer, J., Borgland, S.L., Rho, J.M., & Reimer, R.A. (2021). Prebiotic, probiotic, and synbiotic consumption alter behavioral variables and intestinal permeability and microbiota in *BTBR* mice. *Microorganisms*, 9(9), article number 1833. doi: 10.3390/microorganisms9091833.

[32] Oluwatobi, V.O., Abiola, F.O., & Stephen, O.O. (2024). Unveiling the role of functional foods with emphasis on prebiotics and probiotics in human health: A review. *Journal of Functional Foods*, 119, article number 106337. doi: 10.1016/j.jff.2024.106337.

[33] Read, T., *et al*. (2019). Diversity and co-occurrence pattern analysis of cecal microbiota establishment at the onset of solid feeding in young rabbits. *Frontiers in Microbiology*, 10, article number 973. doi: 10.3389/fmicb.2019.00973.

[34] Shastak, Y., & Pelletier, W. (2023). Review: Vitamin A supply in swine production: Current science and practical considerations. *Applied Animal Science*, 39(5), 289-305. doi: 10.15232/aas.2023-02409.

[35] Shastak, Y., & Pelletier, W. (2024). Review of liquid vitamin A and E formulations in veterinary and livestock production: Applications and perspectives. *Veterinary Sciences*, 11(9), article number 421. doi: 10.3390/vetsci11090421.

[36] Shkromada, O., Suprun, Yu., Fotin, O., Plyuta, L., & Lifar, I. (2024). Determination of the effect of the enzyme and probiotic complex on animal productivity. *Scientific Horizons*, 27(5), 9-19. doi: 10.48077/scihor5.2024.09

[37] Silva, Y.P., Bernardi, A., & Frozza, R.L. (2020). The role of short-chain fatty acids from gut microbiota in gut-brain communication. *Frontiers in Endocrinology*, 11, article number 25. doi: 10.3389/fendo.2020.00025.

[38] Sobri, M., Wiryawan, K.G., Karti, P.D.M.H., & Wibawan, W.T. (2019). Effect of fiber feed on rabbits during the postweaning period. *Pakistan Journal of Nutrition*, 18, 101-108. doi: 10.3923/pjn.2019.101.108.

[39] Song, K., Ling, H., Wang, L., Tian, P., Jin, X., Zhao, J., Chen, W., Wang, G., & Bi, Y. (2024). *Lactobacillus delbrueckii* subsp. *Bulgaricus* alleviates acute injury in hypoxic mice. *Nutrients*, 16(10), article number 1465. doi: 10.3390/nu16101465.

[40] State sanitary norms and rules "Hygienic Requirements for Drinking Water Intended for Human Consumption" (DSanPiN 2.2.4-171-10). (2010, May). Retrieved from https://zakon.rada.gov.ua/laws/show/z0452-10 /.

[41] VNTP-APK-05.07. (2008). *Departmental standards for technological design of enterprises of animal husbandry and rabbit breeding*. Retrieved from https://online.budstandart.com/ua/catalog/doc-page.html?id_doc=67808.

[42] Zhang, Y., Li, Y., Xia, Q., Liu, L., Wu, Z., & Pan, D. (2023). Recent advances of cereal β-glucan on immunity with gut microbiota regulation functions and its intelligent gelling application. *Food Science Nutrition*, 63, 3895-3911. doi: 10.1080/10408398.2021.1995842.

[43] Zhao, C., Li, Y., Wang, H., Solomon, A.I., Wang, S., Dong, X., Song, B., & Ren, Z. (2024). Dietary supplementation with compound microecological preparations: Effects on the production performance and gut microbiota of lactating female rabbits and their litters. *ASM Journals*, 13(1), article number e00067-24. doi: 10.1128/spectrum.00067-24.

# Вплив пребіотику на розвиток лабораторних тварин

**Енджей Ясковський**
Доктор габілітований, професор,
Університет Миколи Коперника
87100, вул. Юрія Гагаріна, 11, м. Торунь, Польща
https://orcid.org/0000-0002-3274-8692
**Василь Лясота**
Доктор ветеринарних наук, професор
Білоцерківський національний аграрний університет,
09117, пл. Соборна, 8/1, м. Біла Церква, Україна
https://orcid.org/0000-0002-2442-2174
**Світлана Ткачук**
Доктор ветеринарних наук, професор
Національний університет біоресурсів і природокористування України
03041, вул. Героїв Оборони, 15, м. Київ, Україна
https://orcid.org/0000-0002-6923-1793
**Надія Богатко**
Доктор ветеринарних наук, професор
Білоцерківський національний аграрний університет
09117, пл. Соборна, 8/1, м. Біла Церква, Україна
https://orcid.org/0000-0002-1566-1026
**Альона Богатко**
Асистент
Білоцерківський національний аграрний університет
09117, пл. Соборна, 8/1, м. Біла Церква, Україна
https://orcid.org/0000-0001-8089-5884

**Анотація.** Пребіотики поліпшують стан здоров'я тварин, але їх позитивна дія на організм залежить від складу та дози препарату, віку та виду тварин. Мета роботи полягала у дослідженні дії на організм білих мишей і кролів нового комплексного пребіотику «Біо-актив». У досліді використовували 40 голів білих лабораторних мишей і кролів. Пребіотик згодовували піддослідним тваринам у дозі 1,2 г/голову. Застосували класичні методики для оцінювання мікроклімату, показників якості водопровідної води для напування тварин, динаміки приростів, гематологічних показників, мікроскопії мазків слизової оболонки сліпої кишки (у кролів). Встановлено, що параметри мікроклімату і показники якості води відповідали вимогам чинних нормативних документів. Згодовування пребіотику білим лабораторним мишам призводило до поступового збільшення маси тіла та середньодобових приростів упродовж всього періоду дослідження. У кролів зростала маса тіла та величина абсолютного і відносного приростів, що сприяло покращенню збереженості та енергії росту тварин. Доведено, що у разі додавання до кормів у годівлі тварин пребіотику збільшувалася кількість еритроцитів, вміст гемоглобіну, загального білка та глобулінів. Виявлено вплив пребіотику на кількість грампозитивних і грамнегативних мікроорганізмів та їх співвідношення у сліпій кишці кролів. При

цьому, кількість грамнегативних мікроорганізмів зменшувалася на 17,7 % ($P < 0,05$), а грампозитивних збільшувалася на 19,4 % ($P < 0,05$). Величина pH вмісту сліпої кишки кролів дослідної групи відповідала значенню 6,5 проти 7,1 у контролі, що свідчило про відновлення функціональної здатності товстого відділу кишечнику. За отриманими результатами дослідження можна констатувати позитивний вплив компонентів пребіотику у дозі 1,2 г/голову на прирости маси тіла, гематологічні показники та кількісний склад мікробіому товстого відділу кишечнику, що важливо для вирощування здорового поголів'я тварин і виробництва якісних харчових продуктів

**Ключові слова:** білі миші; кролі; умови утримання; інтенсивність росту; гематологічні показники; мікроорганізми; сліпа кишка