

Article

# Effect of Innoprovect Probiotics on the Organism of Chickens Vaccinated Against Infectious Bronchitis

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**Abstract:** This article presents data on the study of effective immunoprophylaxis against Infectious Bronchitis in Lohmann Brown breed chicks raised in industrialized poultry farms and smallholder farming operations, through the application of the "Nobilis Ma5 + Nobilis 4/91" Infectious Bronchitis vaccine in combination with the Innoprovect probiotic. Blood samples were collected from all chicks at 7 and 14 days following vaccination and Innoprovect administration, and their immune status was assessed using the Hemagglutination Inhibition (HI) test. A significant increase in antihemagglutinin (antibody) titers was detected in the experimental groups compared to the control group. The stable and elevated maintenance of antibody titers on day 14 of the study indicated the formation of a strong and long-lasting immune response in the chicks. Furthermore, live weight gain in the experimental group was consistently higher than that recorded in the control group.

**Keywords:** Chicks, Infectious Bronchitis Virus (IBV), Vaccine, Innoprovect, Probiotic, Immunity, Immunoprophylaxis, Antigen, Antibody, Antihemagglutinin, Serology.

## Introduction

In recent years, the Government of Uzbekistan has adopted a number of resolutions and regulatory documents aimed at ensuring food security, developing the poultry sector, and meeting the growing demand for poultry products (meat and eggs).

These include the Decree of the President of the Republic of Uzbekistan No. PF-60, dated January 28, 2022, "On the Development Strategy of New Uzbekistan for 2022–2026"; Resolution No. PQ-4015, dated November 13, 2018, "On Additional Measures for the Further Development of Poultry Farming"; Resolution No. PQ-281, dated June 15, 2022, "On Measures to Further Improve the State Support System for the Poultry Industry"; and Presidential Decree No. PF-36, dated February 16, 2024, "On Additional Measures to Ensure Food Security in the Republic," along with other relevant regulatory instruments. These documents serve as important incentives for the implementation of the objectives set forth in this field.

The poultry industry, as a principal branch of agriculture making a substantial contribution to the national economy, is being developed through systematic efforts to increase the number of poultry in limited liability companies, farms, cooperative enterprises, and private subsidiary holdings; to

improve productivity; to enhance the quality of meat and eggs; to ensure proper husbandry practices; and to prevent and treat both infectious and non-infectious diseases, particularly in meat- and egg-type poultry breeds.

The economic losses caused by Infectious Bronchitis and Colibacillosis in poultry — manifested as growth retardation, reduced productivity, and costs associated with treatment and preventive measures — are well documented. The insufficient availability of biological and chemical veterinary preparations further complicates the situation, creating conditions conducive to the more rapid spread of infectious diseases.

In this context, the further development and practical implementation of improved treatment and prophylactic measures against these pathologies represents an urgent challenge. The present research contributes, to a meaningful degree, to the pathomorphological differential diagnosis of Infectious Bronchitis and Colibacillosis in poultry, the development of effective control strategies, and the fulfillment of the objectives established in the relevant regulatory framework.

The poultry sector in the Republic of Uzbekistan is developing rapidly alongside other branches of the national economy. In recent years, infectious diseases such as Infectious Bronchitis and Colibacillosis have emerged as significant constraints on the development and efficiency of commercial poultry production. Addressing these challenges is closely linked to increasing flock numbers, improving productivity, obtaining healthy offspring, ensuring proper husbandry, and protecting birds from disease across smallholder, farm, and private poultry enterprises. The shortage of effective biological and chemical veterinary preparations continues to exacerbate the situation by facilitating the wider spread of infectious diseases, making the development of effective prevention and control measures an urgent scientific and practical priority [1], [2], [3], [4].

Infectious Bronchitis is a highly contagious viral disease widely prevalent in poultry farming, characterized by lesions of the respiratory tract, kidneys, and reproductive system. Research conducted over the past decade has demonstrated that the primary challenge in controlling this disease lies in the high genetic variability of the virus. In particular, a unified phylogenetic classification system based on the S1 gene of IBV has been proposed, revealing that viral strains exhibit an exceptionally broad genetic diversity. This work enabled the evaluation of IBV strains using uniform criteria and established an important theoretical foundation for subsequent molecular-epizootological investigations [5].

The pathogenicity of IBV, its tissue tropism, and serotype characteristics have been experimentally demonstrated to be directly influenced by hypervariable regions within the S1 portion of the spike protein. This study established that mutations in these regions are associated with differences in tissue targeting, severity of clinical signs, and alterations in antigenic properties. These findings indicate that even minor genetic changes in IBV carry substantial epizootological significance in practical terms [6].

In the field of improving vaccination strategies against Infectious Bronchitis, various live attenuated vaccine regimens have been evaluated against IBV variant-2 strains circulating in the Middle East, with certain combination protocols demonstrating considerably greater efficacy in terms of clinical protection and reduction of viral shedding. The principal conclusion drawn from this work is that a single classical vaccine does not yield uniform results across all regions, and that practical vaccination programs must be adapted to locally circulating variants [7].

Regarding the regional evolution of IBV and the continued genetic diversity observed despite vaccination, studies based on Egyptian strains have analyzed the ongoing evolution of IBV, recombination events, and the emergence of novel variants even under active vaccination programs. The authors demonstrate that vaccination programs unsupported by genetic monitoring are insufficient over the long term — a conclusion that is equally relevant to other countries, including regions where the poultry industry is in active development [8].

In the area of rapid and accurate IBV detection, a multiplex real-time RT-PCR assay of considerable practical value has been developed. The assay was designed to differentiate between classical and variant II strains, and the authors demonstrated that such a genotyping approach is valuable for distinguishing between vaccine and field strains — an essential capability for the rapid assessment of epizootological situations and the selection of appropriate preventive measures [9].

Based on our observations and a review of the literature, the implementation of immunoprophylactic measures against Infectious Bronchitis constitutes one of the primary interventions available to specialists, and various immunoprophylactic protocols and approaches are being progressively introduced into production practice.

From the foregoing, it may be concluded that ensuring effective immunoprophylaxis against Infectious Bronchitis in chick rearing and implementing novel immunoprophylactic methods in practical settings represents an urgent priority.

Accordingly, the further refinement and practical implementation of improved preventive measures against this disease – beyond existing general-purpose traditional protocols – remains one of the pressing challenges in the field.

It is in this context that the present research contributes, to a meaningful degree, to the differential diagnosis of Infectious Bronchitis from other infectious diseases, the development of effective control strategies, and the fulfillment of the objectives established in the relevant regulatory framework.

## Materials and Methods

The present study was conducted at "Agro Biznes Qanliko'l" LLC, located in Qonliko'l district, and "Kungrad Golden Egg" LLC, located in Qo'ng'iro't district of the Republic of Karakalpakstan, involving a total of 240 Lohmann Brown Classic layer-type chicks, along with data on the immunoprophylaxis of Infectious Bronchitis implemented at these farms.

Immune response indicators against Infectious Bronchitis in the chicks were determined by the Hemagglutination Inhibition (HI) test at the ELISA and Serology Laboratory of the State Center for Animal Disease Diagnosis and Food Safety of the Republic of Karakalpakstan. Live body weight of the chicks was measured using an A-PLUS model electronic scale.

**Aim of the Study.** To investigate and analyze the efficacy of immunoprophylaxis against Infectious Bronchitis through the application of the "Nobilis Ma5 + Nobilis 4/91" vaccine in combination with the Innoprovect probiotic in Lohmann Brown breed chicks raised under both industrial poultry farming and smallholder farm conditions.

**Objectives of the Study.** To examine the biological characteristics of the vaccination process against Infectious Bronchitis in layer-type chicks. To determine the physiological and immunological effects of the Innoprovect probiotic on the chick organism. To compare the dynamics of live weight gain in experimental and control groups. To evaluate the effect of the Innoprovect probiotic on the level of immunity induced by vaccination against Infectious Bronchitis, through the assessment of antibody titers.

## Results and Discussion

The Innoprovect immunostimulant and the "Nobilis Ma5 + Nobilis 4/91" Infectious Bronchitis vaccine were used in the present study.

We investigated the effect of the Innoprovect immunostimulant on immunity against Infectious Bronchitis under both laboratory and production conditions. Chicks aged 7 and 9 days were vaccinated with the "Nobilis Ma5 + Nobilis 4/91" Infectious Bronchitis vaccine, derived from a strain with a biological activity of  $10^{9.25}$  EL<sub>50</sub>/ml, administered ocularly at a dose of  $10^{4.0}$  EL<sub>50</sub>/ml per bird in a volume of 0.2 ml per head.

A total of 180 Lohmann Brown layer-type chicks sourced from "Agro Biznes Qanliko'l" LLC, located in Qonliko'l district of the Republic of Karakalpakstan, were divided into three groups of 60 birds each. The chicks were housed in KBU-3 cages in the vivarium of the Nukus Branch Laboratory of SAMDVMCHBU and maintained under standardized feeding and microclimatic conditions.

Chicks in the first group received Innoprovect probiotic at a dose of 1 gram per 1 kg of feed. Chicks in the second group received the preparation at the same dose via drinking water. Chicks in the third group served as the untreated control and received no preparation.

Blood samples were collected from all chicks at 7 and 14 days following vaccination and Innoprovect administration, and immune status was assessed using the Hemagglutination Inhibition (HI) test.

The results obtained demonstrated a significant increase in antihemagglutinin (antibody) titers in the experimental groups compared to the control group. This finding indicates that the administration of Innoprovect probiotic activated the immune response of the organism — that is, it potentiated the effect of the vaccine and contributed to a more effective formation of humoral immunity.

In particular, the stable maintenance of elevated antibody titers on day 14 of the study was indicative of the development of a strong and long-lasting immune response in the chicks (Table 1).

**Table 1.** Effect of Innoprovect on the immunogenicity of the vaccine against Infectious Bronchitis.

Groups	Vaccine and method of administration	Number of chicks	Geometric mean antibody titers in blood serum after ... days, in log <sub>2</sub> (M±m)	
			7 day	14 day
I	"Nobilis Ma5 + Nobilis 4/91" ocular + Innoprovect + Innoprovect	60	5,0±0,01	5,4±0,02lk
II	"Nobilis Ma5 + Nobilis 4/91" ocular	60	5,0±0,01	5,8±0,03
III	"Nobilis Ma5 + Nobilis 4/91" ocular (without preparation)	60	5,0±0,01	5,2±0,03

The data presented in the table demonstrate that the administration of Innoprovect probiotic enhanced the immune response in chicks. The highest serum antihemagglutinin titers were observed in Group 2, which received Innoprovect probiotic via drinking water [10].

By day 14 of the experiment, these titers were found to be 0.7 and 0.8 log<sub>2</sub> higher than those recorded in Groups 1 and 2, respectively, and 1 and 2 log<sub>2</sub> higher than those of the control group ( $P < 0.01$ ).

The effect of Innoprovect probiotic in poultry farms was investigated in a comprehensive manner, examining both its influence on the formation of immunity against Infectious Bronchitis and its effect on live weight gain in layer-type chicks.

The study was conducted under conditions approximating production settings, involving 60 chicks sourced from "Kungrad Golden Egg" LLC, located in Qo'ng'iro't district. The chicks were randomly allocated into two equal groups — a control group and an experimental group. No additional preparations were administered to the control group. Chicks in the experimental group received Innoprovect probiotic at a dose of 1 gram per 1 kg of feed on days 3 and 5 of life. Birds in both groups were maintained under standard zooghygienic conditions, with identical feeding, drinking water, microclimatic parameters (temperature, humidity, ventilation), and housing conditions, thereby ensuring the objectivity of the results obtained.

Throughout the experiment, both groups were kept under continuous clinical and laboratory observation, with immunity levels against Infectious Bronchitis monitored by the Hemagglutination Inhibition (HI) test in blood serum [11].

The study revealed a positive effect of Innoprovect probiotic use on the development of immunity against Infectious Bronchitis in chicks (Table 2).

**Table 2.** Results of the study on the strength of immunity against Infectious Bronchitis in chicks under production conditions.

Group	Vaccination and vaccination method	Number of chicks (head)	Geometric mean antibody titers in blood serum after ... days, in log <sub>2</sub> (M±m)	
			7 day	14 day
I Experim ental	"Nobilis Ma5 + Nobilis 4/91" ocular + Innoprovet	30	6,0±0,01	6,0±0,02
	II Control		"Nobilis Ma5 + Nobilis 4/91" ocular	5,0±0,15

Throughout the experiment, the live body weight of the chicks was measured at regular intervals of every 10 days using an A-PLUS model electronic scale [12].

The growth dynamics of the weighed chicks were analyzed in detail (Table 3).

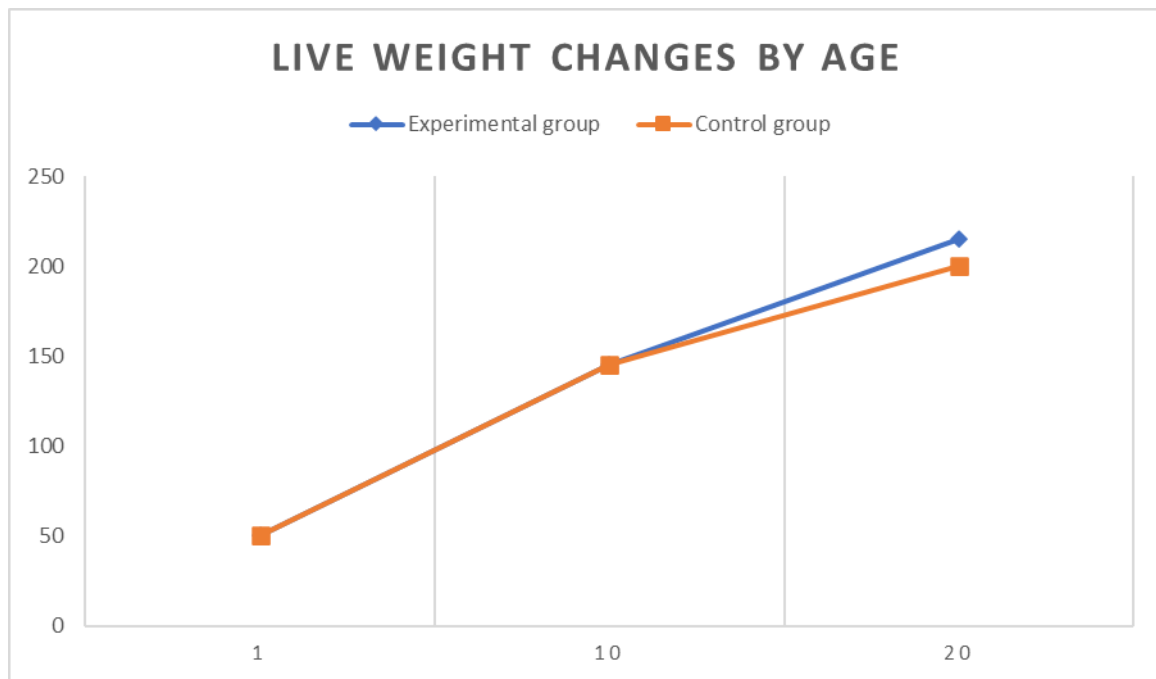
At the start of the experiment, the mean live body weight of one-day-old chicks was identical in both groups, at 50.0 g. Notable differences were observed in subsequent periods. Specifically, at 21 days of age, the mean live body weight of chicks in the experimental group was 168.0 g, compared to approximately 153.0 g in the control group. At 28 days of age, these values were 224.0 g and 205.5 g, respectively [13].

**Table 3.** Live weight of chicks to which Innoprovet probiotic is used.

Age of chicks (day)	Experimental group (g)	Control group (g)
1 day	50.0 g	50.0 g
10 day	168.0 g	153.0 g
20 day	224 g	205,5 g

The results obtained demonstrated that mean weight gain in the chicks increased progressively in a stepwise manner, depending on age, feeding conditions, and the biologically active supplement applied. In particular, under the influence of the Innoprovet immunostimulant administered to the experimental group, metabolic processes were activated, resulting in daily and cumulative weight gain that consistently exceeded the values recorded in the control group. Furthermore, the most intensive periods of growth were identified, during which a marked increase in the live body weight of the chicks was observed. The findings confirmed that mean weight gain in the chicks is directly associated with their healthy development and the proper organization of husbandry technologies [14].

Thus, live weight gain in the experimental group was found to be consistently and stably higher than that of the control group (Figure).



**Figure 1.** Dynamics of live body weight in chicks administered Innoprovect probiotic.

Furthermore, the results of immunological analyses demonstrated that a stronger immune response against Infectious Bronchitis was formed in the group that received Innoprovect probiotic.

In particular, antibody titers in the experimental group were consistently higher, confirming the efficacy of Innoprovect as an immunostimulant [15].

## Conclusion

The results of our study demonstrate that the combined use of the "Nobilis Ma5 + Nobilis 4/91" Infectious Bronchitis vaccine with Innoprovect probiotic significantly enhances the humoral immune response in chicks.

The data obtained on days 7 and 14 of the study revealed a stable and marked increase in antibody titers, confirming the formation of a strong and long-lasting immune response. The highest immunological indicators were recorded in the group that received Innoprovect probiotic via drinking water, highlighting the particular efficacy of this administration route.

Experiments conducted under production conditions corroborated the laboratory findings, demonstrating that antibody titers in the Innoprovect-treated groups were significantly higher than those of the control group — by up to  $2 \log_2$  — thereby providing scientific substantiation for Innoprovect probiotic as an important factor in enhancing vaccine immunogenicity.

In addition, the administration of Innoprovect probiotic exerted a positive effect on the growth and development of the chicks. Live weight gain in the experimental group was consistently higher than that of the control group, indicating a beneficial influence of the preparation not only on the immune system but also on overall metabolic processes.

Our study established the following key scientific and practical findings:

1. Innoprovect probiotic is a safe and effective immunostimulant.
2. Innoprovect probiotic increases vaccine efficacy by 5–8%.
3. Innoprovect probiotic improves live weight gain by an average of 7.8%.
4. Innoprovect probiotic is recommended as a promising agent for enhancing immunoprophylaxis against Infectious Bronchitis.

The introduction of Innoprovect probiotic into poultry production practice will improve the effectiveness of control measures against Infectious Bronchitis, reduce economic losses, and contribute to increased productivity. For these reasons, we consider the widespread application of Innoprovect probiotic in both industrial and private poultry farming enterprises to be highly advisable.

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