



PAPER ID: 10A15J



## ANTIGENIC ACTIVITY OF AN EXPERIMENTAL INACTIVATED VACCINE AGAINST CHICKEN INFECTIOUS BRONCHITIS

Edward Javadov<sup>a\*</sup>, Oleg Khokhlachev<sup>b</sup>, Olga Kozyrenko<sup>a</sup>, Irina Vikhreva<sup>b</sup>, Olga Polyakova<sup>a</sup>

<sup>a</sup> Department of Epizootology V.P. Urbana of St. Petersburg State Academy of Veterinary Medicine, RUSSIA

<sup>b</sup> Research Consulting and Diagnostic Center for Poultry of "St. Petersburg Academy of Veterinary Medicine", RUSSIA.

### ARTICLE INFO

#### Article history:

Received 25 June 2019  
Received in revised form 31 July 2019  
Accepted 09 August 2019  
Available online 26 August 2019

#### Keywords:

Infectious bronchitis of hens; Field isolate of the IBC virus; Veterinary; Inactivated vaccine against IBC; Antigenic activity; Vaccinated chicken.

### ABSTRACT

The article presents materials on the study of the antigenic activity of an experimental inactivated emulsion vaccine based on the field isolate of the chicken infectious bronchitis virus isolate that is identical to the variant strain QX of the IBC virus. The research results allow us to consider the obtained virus isolate as a candidate for the manufacture of inactivated vaccines against infectious bronchitis of hens.

This article is published as part of research work by the federal state budgetary educational institution of higher education "St. Petersburg State Academy of Veterinary Medicine" by order of the Ministry of Agriculture of Russia at the expense of the federal budget in 2019. Subject: Study of the circulation of variant strains of the chicken infectious bronchitis virus in poultry farms of the Russian Federation.

©2019 INT TRANS J ENG MANAG SCI TECH.

## 1. INTRODUCTION

Infectious bronchitis of chickens (IBS) still poses a real threat to industrial poultry farming, despite intensive programs for the specific prevention of the disease (Kuklenkova, et al., 2018). This is due to the identification in different regions of the world, including in the Russian Federation, field IBA virus strains that differ in virulence, antigenic and immunogenic activity: D274, D1466, 793B, CR88, 4/91, IT02, D388, QX, etc. (Abdullov, 2015). The emergence of new variant strains of the virus is due to the genetic structure of coronavirus IBC, amenable to relatively rapid mutation and genetic recombination. Circulation in poultry farms of virulent field, incl. variant strains of the IBC virus make it extremely difficult for veterinarians to diagnose, prevent and control the disease (Shcherbakova, et al., 2018).). The circulation of variant strains in poultry farms of the Russian Federation, in addition to the case of poultry, leads to a significant decrease in egg productivity and the quality of the resulting eggs see Figure 1.



**Figure 1:** Eggs obtained from laying hens with chicken infectious bronchitis

Widely used in previous years, live and inactivated vaccines based on the classic Massachusetts serotype virus from strains H-120, H-52, Ma5, M41, Chapaevsky currently in many cases do not provide the formation of reliable immunity in vaccinated birds (Kuklenkova, et al.,2018).). This requires new approaches to the study of the epizootic situation in each individual region and the poultry industry. A significant role in this work is played by comprehensive laboratory studies of the material, aimed at isolating the circulating virus, determining its species and strain affiliation. The results of the analyzes contribute to the development of effective programs for the specific prevention of IBS, based on the use of autogenous vaccines prepared on the basis of field virus isolates (Balykina, et al., 2018). The objective of the present studies was to study in an experiment on chickens the antigenic activity of an experimental inactivated emulsion vaccine prepared on the basis of a field isolate of the causative agent of the IBI virus, identical to the variant strain QX of the IBC virus (Javadov, et al., 2008). The data obtained can be used to obtain the inactivated IBA virus antigen and its possible use in the manufacture of mono- and multivalent inactivated vaccines against this disease.

## **2. MATERIALS AND METHOD**

### **2.1 VIRUS REPRODUCTION**

For the manufacture of the vaccine, a field isolate of the IBC virus was used. Reproduction of the virus in order to obtain a brood of viral material was performed on developing SPF embryos of 9-day incubation chickens (Lohmann Tierzucht, Germany). The infectious activity of the obtained virus was checked on SPF chicken embryos (Teryukhanov, 1976). Titration was performed using the virus in successive 10-fold dilutions. The virus was pre-checked for the absence of bacterial contamination by plating on culture media. The calculation of the titer of the infectious activity of the virus was carried out according to the method of Reed and Mench.

### **2.2 VIRUS INACTIVATION**

The virus was inactivated using the domestic chemical preparation Ecodezrico, containing

1,8,3,6-diendomethylene-1,3,6,8-tetraazacyclodecane sodium hydrogen phosphate as the active substance, in a final concentration of 0.12%. When conducting inactivation, we used separate methodological materials described in the article by D. Glazer et al. Upon completion of inactivation, the quality of the virus inactivation was monitored by the method of successive passages of inactivated viral material on 9-day-old incubation hens SPF embryos. For this purpose, inactivated viral material in the volume of 0.2 cm<sup>3</sup> was injected into the allantoic cavity in each embryo, followed by incubation in the prescribed mode. The safety of embryos and the presence of pathological signs characteristic of IBI were taken into account. Then, allantoic fluid was taken from each embryo, which was used for the second passage. Then spent the final third passage.

### **2.3 GETTING VACCINE EMULSION**

For the preparation of the vaccine in the form of an inverse emulsion "water in oil" used ready-made oil adjuvant Montanide ISA 70vc (Seppic, France). Emulsification was performed using an Ultra-Turrax T-25 apparatus.

### **2.4 EXPERIMENTAL BIRD**

To study the antigenic activity of the experimental inactivated vaccine, 35 heads of 10-day-old broiler chickens of the Ross-308 cross were used, obtained from a farm that is safe for respiratory diseases of birds. At the beginning of the experiment, there were no antibodies to the IBI virus in the blood serum of chickens. The chickens were kept in a cage. Conditions of feeding and feeding (the composition of the diet, temperature regime, and illumination in the boxes of the vivarium, basically, corresponded to the hygiene standards for birds of this cross-country and age). Chickens ringed. 2 groups of chickens were formed: 1 group (10 goals) - intact chickens ("clean" control); Group 2 (25 goals), chickens, each of which was administered an inactivated vaccine subcutaneously, in the lower third of the back of the neck in a volume of 0.5 cm<sup>3</sup>. The bird was monitored for 35 days after immunization (observation period). The safety and clinical condition of the birds was recorded daily. Blood was taken from control and immunized birds before vaccination and 21-, 28-, and 35 days after vaccination. The resulting serum was examined for the presence of antibodies to the IBI virus.

### **2.5 LINKED IMMUNOSORBENT ASSAY**

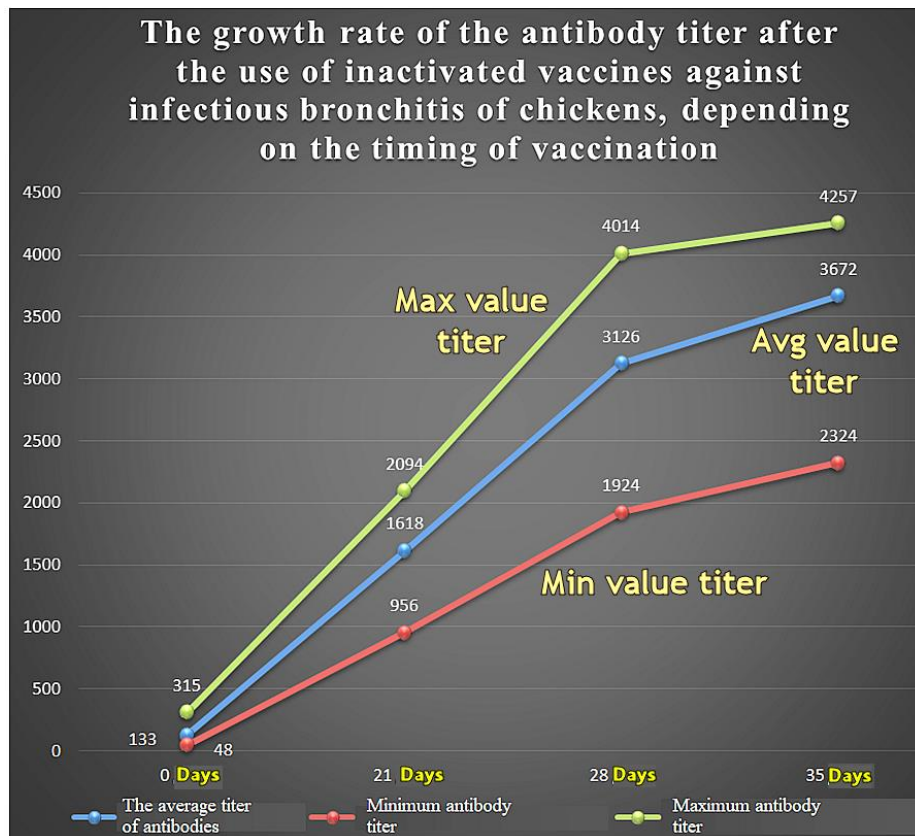
Antibodies to IBA virus in the blood serum of the experimental bird were detected by enzyme-linked immunosorbent assay (ELISA, ELISA) in an indirect solid-state version of the reaction. We used IDEXX test systems and the x-Chek computer program. ELISA results were recorded on a Sunrise TECAN spectrophotometer.

## **3. RESULTS AND DISCUSSION**

When the virus was titrated on chicken SPF embryos, the titer of the infectious activity of the obtained virus was established, which amounted to 7.5lg EID<sub>50</sub>/cm<sup>3</sup>.

The control of the inactivated virus for completeness of inactivation revealed a complete and irreversible loss of the infectious properties of the virus.

The results of the study in ELISA of blood serum samples of vaccinated chickens for the presence of antibodies to the IBI virus are presented in Figure 2.



**Figure 2:** Antigenic activity of inactivated vaccine against IBS at different times after immunization (23 pieces of the sample).

It was found that the inactivated emulsion vaccine has a pronounced antigenic activity and causes vaccinated chickens to produce antibodies to the IBI virus in high protective titers. In chickens in the “pure” control group, no antibodies to IBI virus were detected in positive diagnostic titers in any case. The death of the bird during the experiment and the increased post-vaccination reaction of local and / or general nature was not noted.

#### 4. CONCLUSION

An experimental inactivated emulsion vaccine based on the isolate of the chicken infectious bronchitis virus isolate, identical to the variant strain QX of the IBC virus, provides the formation of humoral immunity in vaccinated chickens in protective titers after a single immunization.

#### 5. AVAILABILITY OF DATA AND MATERIAL

Data can be made available by contacting the corresponding author.

#### 6. ACKNOWLEDGEMENT

This article is published as part of research work by the Federal State Budgetary Educational Institution of Higher Education "St. Petersburg State Academy of Veterinary Medicine" commissioned by the Ministry of Agriculture of the Russian Federation with financial support of the 2019 federal budget, Research Topic: Studying the circulation of variant strains of the chicken infectious bronchitis virus in poultry farms of the Russian Federation.

#### 7. REFERENCES

Javadov, E. D., Khokhlachev, O.F., et al. (2008). *Infectious bronchitis of chickens is the cause of respiratory syndrome in broiler chickens*. In the press.

- Glaser, D. A., Frolov, C. V., Borisov, A. V., Kulakov, V. Yu. (2008). Obtaining inactivated chicken infectious bronchitis virus (strain "Kaluga"). *Veterinary consultant*. 20(183), 3-4.
- Abdulloev, H. S., (2015). *Immunobiological properties of the infectious bronchitis virus of chickens of the QX genotype*. Abstract of diss. Cand. veterinary sciences. Federal Center for Animal Health. Vladimir.
- Shcherbakova, L. O., Kolosov, S. N., Nikonova, Z. B., Ovchinnikova, U. V., Praise, I. A. (2018). Genetic characteristics of hepatitis B virus isolates detected in the CIS countries in 2015-2017. *Veterinary medicine today*. 3(26), 30-34.
- Teryukhanov, A. B. (1976). *Infectious bronchitis of chickens*. L., "Kolos" (Leningrad Branch). 44-45.
- Kuklenkova, I. V., Zhavoronkova, T. S., Pashkin, A. V., Sochnev, V. V., Pashkina, Yu. V., Kozyrenko, O. V., Usenkov, A. V., Filippov, N. V., Gusev, A. A., (2018). Optimization of conditions for the production of viral antigen of infectious bronchitis of chickens for the manufacture of inactivated vaccines. *Issues of regulatory regulation in veterinary medicine*. 4, 47-50.
- Kuklenkova, I. V., Samodelkin, A. G., Pashkin, A. V. C., Avilov, V. M., Sochnev, V. V., Kozyrenko, O. V., Usenkov, A. V., (2018). Immunogenic activity of the viral polyvalent inactivated vaccine against the Newcastle disease, infectious bronchitis, egg drop syndrome, reoviral and metapneumoviral infection in poultry. *International Journal of Pharmaceutical Research*. 10(4), 675-679.
- Balykina, A. B., Nikonov, I. N., Karpenko, L. Yu., Bakhta, A. A., Kuznetsov, Yu. E., (2018). Influence of the animal feed components and biologically active substances into the intestinal microbiota state of the bird. *Research Journal of Pharmaceutical, Biological, and Chemical Sciences*. 9(6), 1876-1880.
- Balykina, A. B., Nikonov, I. N., Karpenko, L. Yu., Bakhta, A. A., Kuznetsov, Yu. E. (2018). The composition and role of the microbiota of chickens gastrointestinal tract. *Research Journal of Pharmaceutical, Biological, and Chemical Sciences*. 9(6). 1881-1885.



**Professor Dr. Javadov Eduard** is Professor of Epizootology V.P. Urbana of St. Petersburg State Academy of Veterinary Medicine, RUSSIA. He graduated from Leningrad Veterinary Institute. He holds a Doctor of Veterinary Sciences degree. He is an Academician of the Russian Academy of Sciences (2016). His researches are Immunobiological Monitoring, Mono- And Bivalent Inactivated Vaccines Against Highly Pathogenic Avian Influenza, Infectious Bursal Disease, and Avikron.



**Oleg Khokhlachev** is a Leading Specialist of the Scientific Research Consulting and Diagnostic Center for Poultry of "St. Petersburg Academy of Veterinary Medicine". He is a Candidate of Veterinary Sciences. His research encompasses Poultry Sciences.



**Dr. Kozyrenko Olga** is an Associate Professor and Head of the Department of Epizootology V.P. Urbana, St. Petersburg Academy of Veterinary Medicine, Russia. She holds a Doctor of Veterinary Sciences degree. Her research is related Biological Hazards and Veterinary Science.



**Irina Vikhreva** is a Leading Specialist of the Research Consulting and Diagnostic Center for Poultry of the St. Petersburg Academy Veterinary Medicine, Russia. She is interested in Poultry Studies



**Polyakova Olga** is an Associate Professor at the Department of Epizootology named after V.P. Urbana, St. Petersburg Academy of Veterinary Medicine. She is a Candidate of Veterinary Sciences. Her research involves Modern Poultry Farming.