Khazar Journal of Science and Technology Volume 3 №1 2019, 53-57

DOI: 10.5782/2520-6133.2019.3.1.53

Sensitivity of Primary Tripsinized Cell Systems EYQ and FEC to the Fowl Pox Virus

Kubra Yusifova
Veterinary Scientific Research Institute, Azerbaijan
Corresponding author: k.kubra@list.ru

Abstract

Viral diseases like fowl pox create serious problems for industrial poultry. Fowl pox is a disease found in poultry worldwide and is caused by viruses of the family Poxviridae and the genus Avipoxvirus. The viruses causing fowl pox are distinct from each other but are antigenically similar. Possible hosts include chickens, turkeys, quail, canaries, pigeons and many other species of birds. It is known that the cultivation of a fowl pox virus in chicken embryo cell cultures is common practice for vaccine production. Many scientists use different local strains of fowl pox virus in attempts to improve the cultural vaccine against chicken fowl pox. The work described below demonstrates the possibility of cultivating and grouting a "Baku" strain of fowl pox virus in a primary cell culture of Japanese quail embryos.

It was discovered that the "Baku" bird strain of fowl pox virus has adapted to both cell systems. An increase in the titer of fowl pox virus was observed in both Japanese quail embryo cell culture and in that of chicken fibroblast embryos. It was found that the culture of Japanese quail embryo cells is a more effective tissue culture for vaccine production than chicken embryo cell culture because of its simplicity, economy, absence of extraneous contaminants and its stable biological properties. It was established that a cell culture of Japanese quail embryos is a promising basis for the creation of highly immunogenic specific prophylaxis against avian fowl pox.

Keywords: fowl pox, virus, primary cell culture, strains virus, vaccine

Introduction

Viral diseases like fowl pox create serious problems for industrial poultry. It is widespread and causes significant economic damage to poultry farms. It is already

known that the cultivation of a fowl pox virus in chicken embryo cell cultures is common practice in preparing biomass for vaccine production. It is also known that the fowl pox virus has a cytopathic effect in cell culture, and with prolonged passage it becomes less contagious to the bird, but retains immunogenic properties. Last year's anti-bird fowl pox cultural vaccines are not inferior in their immunogenicity to embryonic preparations (Patrushev, 1967; Sukhorukov, 2012; Zenov, 2010). Designed in Azerbaijan's VCRI by F. B. Shirinov and A. N. Gojayev in 1985, the embryonic anti-chicken fowl pox vaccine from the "Baku" strain was grown on chicken embryos, and was thus imperfect in its production (Cherkezova, 2003; Elizbarashvili, 1987). The disadvantage of embryonic anti-bird fowl pox viral vaccines is the low level of infectious activity in chickens (Syurin, 1956; Tarasenko, 2003). In these circumstances, the search for fowl pox virus strains in birds adapted in a cell culture with highly infectious and immunogenic properties is appropriate for the production of modern drugs. Determining the sensitivity of primary cell cultures to fowl pox virus, monitoring the dynamics of the accumulation of poxvirus in a cell culture is appropriate in modern veterinary medicine.

In some countries, vaccines with attenuated virus strains are used in disease prevention: "Intervet" in the Netherlands, "Webster" in Australia, and others (Patrushev, 1967). Attenuated anti-bird fowl pox vaccines have been registered in Azerbaijan since 2002 to the present: in 2002 "Nobilis AE+Pox", by the Dutch company Intervet International BV; in 2009 - "AviPro AE-Pox" and "AviPro Pox", by the German company Lohman Animal Health International; in 2011 - "Gallivac AE+FP", by the French company Merial; in 2014 "Avivak-fowl pox", by the Russian company Avivak NPP (Silva, 2009).

The "AviPro Pox" vaccine is made from chicken embryos infected with bird fowl pox viruses, the "HP-B" strain. The "FPC" strain is used in the production of the German vaccine "AviPro AE+Pox" by the Lohman Animal Health Gmb @Ko KG company.

For the vaccine "Nobilis AE+Pox" the company MSD Animal Health, or Intervet (Holland), uses the "Gibbs" strain. This strain was used also for the "Pigeon Pox" and "AVA+Pox+CE" vaccines (Silva, 2009).

The embryo anti-bird fowl pox vaccine "Avivak Ospa" is made from a live attenuated strain of fowl pox virus, "K" strain. The "Cutter" strain is widely used in the production of the associated vaccines "Gallivac AE+FP" by the Merial company. The "Hitchner", "Calnek" and "Cutter" strains are used in the production of "Pox blen" associated vaccines. The "Cutter" Strain is also used by the Ceva Sante Animale company in the production of the associated vaccines "Cevac FP+L",

“Cevac Poximune”, “Cevac Poximune® AE”, “Cevac Vectormune FP+MG”, “Cevac Vectormune FP+MG+AE”, “Cevac Vectormune® FP+N+A” and “Cevac Vectormune® FP+LT+AE”. The Lohmann Animal Health INT company uses “P” strains for its “AviPro Pigeon Pox P” vaccine. For the “AviPro Pigeon Pox C” vaccine, it uses “C” strain. The “Chicken PV” strain is used for the vaccines “AviPro Pox Ceo”, “AviPro Pox TC”, “AviPro Pox” and “AviPro AE+Pox+TC”. Russian anti-bird fowl pox vaccines include the strains "K", "Ospovak", "27-ASh" and "ND" (Meseko, 2012; Yusifova, 2016; Yusifova, 2015). So, researchers currently use mainly local strains to improve their vaccines. This is important for increased effectiveness of vaccination against fowl pox.

The study in this article is aimed at a comparative analysis of the “Baku” strain of avian pox virus adapted to the culture of quail embryo cells and chicken fibroblast cell culture to obtain a highly pathogenic antigen.

To this end we conducted successive passages of fowl pox virus on primary cell systems to preserve and increase its antigenic properties.

Materials and methods

Cell cultures were prepared by the standard trypsinization method. The sensitivity of the cell culture was determined by infecting it with bird fowl pox virus in dilutions of up to 10^{-4}. Changes in cell culture were detected by microscopy (Syurin, 1956). During cultivation, the timing of the onset and the nature of the cytopathic effect were determined. The titer was determined by the Reed and Mench method, ($EID_{50/ml}$), and in the RHA reaction, the virus titer was respectively expressed in $HAU_{50/0,5мл}$. (Yusifova, 2016; Yusifova, 2015).

Results and discussion

We performed 25 passages of fowl pox virus on cell cultures of Japanese quail embryos and chicken fibroblasts. The biological properties at the time of adsorption of fowl pox viruses in the primary cell systems of Japanese quail embryos and chicken fibroblasts were studied, including the optimal conditions for their reproduction in the systems studied. The destruction of the primary cell culture under the influence of fowl pox virus was observed by microscopy.

Preliminary studies have shown that the cytopathic phenomenon in cell cultures of chicken fibroblast embryos can occur in the first stages of fowl pox virus infection. So, the cytopathic effect in the culture of chicken fibroblast embryo cells was observed in passage 2-3, in the culture of Japanese quail embryo cells in passage 10

on day 5 after infection. In both cell cultures, cell destruction was observed in the first 48 hours, the integrity of the monolayer was not violated. Monolayer destruction was observed in the next 72 hours, and evaluated «+ +» and «+ + +». The cytopathic effect of viruses is equally evident in chicken embryo cell culture and in Japan quail cell culture. But the onset time of the cytopathic effect in the cell cultures was not the same. The cytopathic effect of the fowl pox virus “Baku” strain in the culture of chicken embryo cells was observed 96 hours after infection, but in the culture of quail embryo cells 144 hours after infection.

Conclusion

The results of the studies showed that the fowl pox virus "Baku" strain adapted to both cell systems. We observed an increase in the titer of fowl pox virus in both the culture of Japanese quail embryo cells and in the culture of chicken fibroblast embryos. In both cultures, the virus titer was 256 -512 ГАЕ/$_{0,5ml}$. It was found that the culture of Japanese quail embryo cells is a more effective tissue culture for vaccine production than chicken embryo cell culture because of its simplicity, economy, absence of extraneous contaminants and its stable biological properties.

It was established that a cell culture of Japanese quail embryos is a promising system for creating highly immunogenic specific prophylaxis against avian fowl pox.

References

Cherkezova, T.V. (2003). Materials of the All-Union Scientific Conference “Formation of immunity to fowl pox in young chickens” p.11- 12, Moscow.

Elizbarashvili, E.I. (1987). Features of the cultivation of fowl pox viruses in birds in a growing and transplanted tissue culture", p.13-15, Moscow.

Meseko, C.A., Shittu I.A., & Akinyede O. (2012). Seroprevalence of Fowl Pox Antibody in Indigenous Chickens in Jos North and South Council Areas of Plateau State, Nigeria: Implication for Vector Vaccine. p.4.

Patrushev, F.M. (1967). The study of some biological properties of fowl pox viruses in birds." Abstract, Perm, p.10-14.

Silva, P.S., Batinga, T.B., & Sales, T.S. (2009). Fowl pox: identification and adoption of prophylactic measures in backyard chickens in Bahia, Brazil; Rev. Bras. Campinas Cienc. Avic. vol.11 no.2, p.4.

Sukhorukov, O.A. (2012). Physiological characteristics of quail and ways of correcting the functional state and production indicators, Tomsk Agricultural Institute - Branch of FSEI HPE Novosibirsk State Agrarian University, Russia, p.3-5.

Syurin, V.N. (1956). Belousova R.V., Fomina N.V. Veterinary Virology.1956.

Tarasenko, E.V. (2003). Comparative immunobiological characteristics of vaccine strains of fowl pox viruses of birds adapted to cell culture», p.4-7, Moscow.

Yusifova, K.Y. (2016). Optimal Method of Influenza Virus Infection in the Cell. A: The "agrarian". № 5, p.25-26, Moscow.

Yusifova, K.Y. (2015). Biological characteristics of avian pox viruses in cell culture. Proceedings of the II International Conference of the Young Scientists' Council of the Academy of Agrarian Sciences of the MAA. p.149, Baku.

Zenov, N.I. 2010. Quail embryos - the basis for the production of virus vaccines against Marek's disease and viral diseases in children - measles and mumps, p.2-4.