

Duration of immunity following the administration of oil-based avian influenza H₅N₁ vaccine in layers

Maryam NISAR, Anwar-ul-HAQ, Asif RASHID*, Muhammad IQBAL
Veterinary Research Institute, Lahore Cantt 54810 - PAKISTAN

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Abstract: Avian influenza (AI) occurs worldwide and causes tremendous economic losses. The disease is characterised by respiratory signs, depression, and reduced food and water intake. In the present study, an oil-based vaccine was prepared by using Montanide ISA 70 MVG, was prepared and the duration of immunity checked at different time intervals. For this purpose, the cumulative mean titre (CMT) was calculated after employing haemagglutination inhibition test in 50 pullets at day zero before vaccination and then inoculated 0.3 mL of emulsified vaccine subcutaneously in 40 birds while 10 birds were kept non-vaccinated as a control group throughout the experiment. On day 21, a booster dose of 0.5 mL emulsified AI vaccine was injected into the vaccinated birds that produced highest CMT at day 63 and maintained the protective antibody titre on day 84, 105, 126, 152, 182, and 228 in layers. The study showed that a booster dose at day 21 produced a high protective AI antibody titre in birds that minimises the stress of repeated shots of aqueous vaccine.

Key words: Avian influenza, Montanide ISA 70 MVG, cumulative mean titre, haemagglutination inhibition test, layers

Introduction

Avian influenza is an infectious viral disease that affects poultry, caused by type A influenza virus belonging to the family orthomyxoviridae, genus influenza virus A. The disease is characterised by respiratory signs, depression, and reduced feed and water intake. In egg laying birds, there is a decline in egg production and quality (1). The financial losses due to an epidemic of avian influenza can be huge and management of outbreaks by stamping policy and pre-emptive culling policy alone leads to very high cost and economic losses for the public sector, the industry and ultimately for consumers (2).

In Pakistan, the disease was first reported in breeder flocks around Rawalpindi/Islamabad during November 1994 and caused heavy losses up to mid 1995 (3). Avian influenza is an important zoonotic disease. In addition to the poultry outbreaks, 241 human infections with H₅N₁ were confirmed, out of which 141 were fatal as reported by the World Health Organization (4).

Vaccination is an effective way of controlling infectious diseases and has achieved spectacular success in promoting human and animal welfare (5). During the outbreak of bird flu disease in northern areas of Pakistan, the Veterinary Research

* E-mail: dr_asif_rasheed@hotmail.com

Institute in Lahore prepared an aqueous vaccine on a war footing basis that effectively controlled the disease. However, due to quick absorption and shorter duration of immunity by aqueous vaccine, there was a need to prepare an oil-based vaccine that would enhance immunity and minimise the stress caused to the birds by repeated shots of aqueous vaccine.

In the present study, the duration of immunity against avian influenza (H₅N₁) disease was reported at different time intervals following the administration of oil-based (Montanide ISA 70 MVG) avian influenza vaccine in birds.

Materials and methods

Virus and vaccine

Avian influenza virus sub-type H₅N₁ (A/Chicken/NARC/2238/06) was obtained from the Ministry of Forestry, Agriculture and Livestock (MINFAL) in Islamabad, Pakistan. The virus was propagated on 10-day-old embryonated chicken eggs. Chorioallantoic fluid (CAF) was harvested and formalised. One batch of avian influenza oil-based vaccine was prepared after conducting all quality tests as described in OIE terrestrial manual (6).

Experimental design

Fifty pullets of 6 weeks of age were procured from the poultry unit section of the Veterinary Research Institute in Lahore. Emulsified vaccine (0.3 mL) was inoculated subcutaneously to 40 pullets while 10 birds were kept as non-vaccinated controls in a separate compartment. A booster dose of 0.5 mL was given to all vaccinated birds after 3 weeks. All birds were kept under observation for 228 days and provided with balanced feed.

Serology

Before vaccination, antibody titre was checked against avian influenza virus. After vaccination, serum samples were collected on days 21, 42, 63, 84, 105, 126, 152, 182, and 228 and cumulative mean titre (CMT) were calculated by employing a haemagglutination inhibition (HI) test (7).

Results

In process of vaccine

The safety and sterility of prepared avian influenza oil-based vaccine were satisfactory. All the birds remained apparently healthy without any adverse reaction and signs of illness from the immunisation oil-based AI vaccine.

Serology

On day zero, all 40 birds were inoculated with oil-based avian influenza at the dose rate of 0.3 mL subcutaneously. The cumulative mean titre (CMT) was calculated after employing a HI test; it was 3.56 on day 21.

A booster dose of 0.5 mL was injected into the vaccinated birds on day 21 and CMT was calculated on day 42, 63, 84, 105, 126, 152, 182, and 228; the results were 8.25, 8.82, 7.12, 6.86, 6.13, 5.92, 5.86, and 5.73, respectively. After the booster dose, CMT was increased on day 42 and it was at its peak on day 63. After that CMT was gradually decreased and on day 228 it was 5.73 (Figure).

Discussion

Vaccination is part and parcel of good husbandry and management. In the present study, the duration of immunity against oil-based avian influenza (H₅N₁) vaccine was reported. Numerous experimental studies using vaccine against H₅N₁ highly pathogenic avian influenza (HPAI) have demonstrated the value

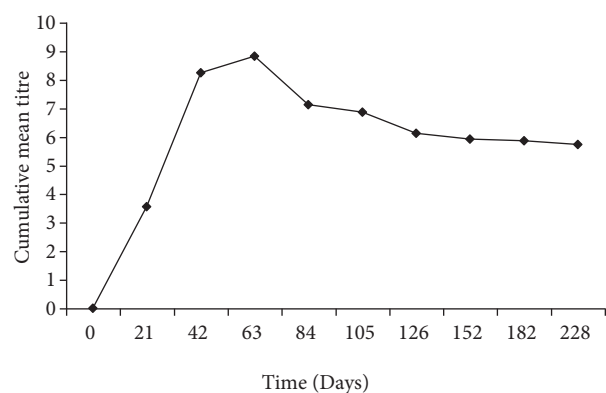


Figure. Cumulative mean titre (CMT) by haemagglutination inhibition test for the duration of avian influenza H₅N₁ vaccine at different time intervals.

of vaccination in protecting poultry from disease in minimising virus excretion (8,9).

There is limited published literature regarding the duration of oil-based avian influenza vaccine in birds. The present study has revealed the protection vaccination gives against avian influenza disease. A similar study was conducted by Stone (10) that used 0.5 mL of vaccine and concluded that there was 90%-100% protection in the vaccinated broiler against death and morbidity. The same author also reported maximum geometric mean titre (GMT) in layers i.e. 1:597 by 2 consecutive doses of vaccines in layers and 1:422 by 1 dose. This study is in agreement with the

present study, which concluded that a booster dose of oil-vaccine increased the duration of immunity. Similarly, Hwang et al. (11) immunized chickens with oil-adjuvant inactivated H5N1 vaccine, which protected from lethal infection by highly pathogenic H5N1 influenza virus until 12 weeks.

In conclusion, a booster dose of Montanide ISA 70 MVG based vaccine on day 21 produced high protective antibody titre in birds, which maintained the protective antibody titre for long interval in layers and reduced the stress of repeated shots of aqueous vaccine in breeder flock.

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