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**Research Article** 

# Determination of Immune Response of Cevac New L® (La Sota Strain) Vaccine in Pigeon

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#### Abstract

Newcastle disease is a devastating poultry disease all over the world including Bangladesh. The present study was conducted to investigate the immune response of ND live vaccines Cevac New L® (La Sota strain) in pigeons during the period from January, 2014 to June, 2014. For the purposes of this experiment 40 pigeons were divided into 2 groups. In group A (n=20) and B (n=20,unvaccinated control) vaccination was performed twice with Cevac New L® at day 5 and 21 of age i/o. Serum antibody titre was evaluated by the means of hemagglutination inhibition test (HI). The Mean $\pm$ SD of HI titre (log2X) of group A was 4.75 $\pm$ 0.64, 5.65 $\pm$ 0.59, 6.85 $\pm$ 0.67, 7.55 $\pm$ 0.51,6.35 $\pm$ 0.67, 5.20 $\pm$ 0.77 at day12, 19, 28, 35, 42 and 49 of post vaccination respectively. It was observed that serum Ab level was gradually increased from day 12 to day 35 and declined slowly after day 42 in group, leading to good protection to the pigeon against ND. There was a significant (p<0.05) differences between the HI titre of group A and B. From the result, it was revealed that Cevac New L® (La Sota Strain) produced better immune response in the pigeon body against ND in respect of HI Abtitres response.

## **INTRDUCTION**

Newcastle disease is a highly contagious, rapid spreading nature and World Organization for Animal Health (OIE) listed notifiableviral disease affecting all poultry species, characterized by gastrointestinal, respiratory and nervous signs with high mortality (up to 100 %) [1]. ND is a major problem to commercial poultry industry throughout the world [2]. Epizootics of ND in poultry continue to occur on a regular basis in Africa, Asia and Central and South America while sporadic epizootics occur in Europe [3]. In Bangladesh ND is mostly due to velogenic strain [4]. During 1981 to 1985, infections of racing and show pigeons with an APMV-1 became worldwide, causing a frequently fatal disease primarily associated with neurological signs. Newcastle disease is endemic in Bangladesh with prevalence of viscerotrophic velogenic strain [5]. Effective control of ND relies on the use of safe and good vaccines. Live vaccines prepared with lentogenic and mesogenic strain of NDV are now more commonly used in pigeon. This is because live vaccines can be produced on a large scale at a relatively low cost. The vaccines are easy to administer on a large scale and rapidly stimulate humoral, cell-mediated and mucosal surface immunity in the vaccinated birds. Vaccination as directed by [4]. includes administration of live lentogenic vaccine (BCRDV) of F-strain by inta-ocular inoculation followed by a live mesogenic vaccine (RDV) of mukteswar strain by intramuscular route maintenance of sustainable level of neutralizing antibody in the serum of vaccinated birds. Various live vaccines containing lentogenic strains of NDV such as  $B_1$ , La Sota, VG/GA and ND clone are brought by several importers. The best-known lentogenic live vaccines are Hitchner  $B_1$  and La Sota strain, both of which hadbeen extensively used [6].

Newcastle disease vaccines produced in Bangladesh are not sufficient to meet up the demand of growing poultry industry. As a result a good number of live lentogenic and mesogenic as well as inactivated Newcastle disease vaccines are being imported and marketed. Vaccination against ND in poultry is very common in Bangladesh. But in pigeon vaccination against ND is very rare because of lack of proper scientific knowledge regarding vaccine and vaccination against ND. In most of the cases, the vaccines were found effective against ND incase of chicken. Although there was no study found regarding immune response of ND vaccines in pigeon except Vaccination of birds either killed or live NDV vaccines might induced cell mediated humoral local and passive immunity. Antibody titers against NDV were usually evaluated by means of hemagglutination inhibition test (HI) was widely used including Bangladesh. That's why research aims to study the assessment of NDV "Lasota strain" vaccines (Cevac New L®) induced immune response determined by HI test.

#### **MATERIALS AND METHODS**

The study was carried out in experimental shed of department of Medicine and the laboratory of Microbiology and Immunology, Sylhet Agricultural University, Sylhet, during the period of January 2014 to July 2014. Pigeons were collected from the local market of Sylhet with no history of vaccination against ND and carried to the experimental shed. The pigeon were reared under strict bio-security and prerequisite required for rearing such birds. All pigeons were supplied with nutritional feed and fresh drinking water through the entire experiment. Among all pigeons, 40 pigeons were grouped into A and B and the pigeons of group A were vaccinated with Cevec New L® at day 5 and day 21 respectively while pigeons of B group were kept as unvaccinated control. Blood was collected individually from the brachial vein at day 4, 12, 19, 28, 35, 42 and 49 with the sterile syringe and needle and placed the syringe in a slanting position for 1 hour at room temperature. Then the clot was detached from the wall of the syringe carefully, allowed it to settle down. Serum was then collected carefully and aseptically, transferred into small vials and then centrifuged at 1500 rpm for 15 min to obtain clear serum were stored at-20C temperature until used [7]. Antibody titer for NDV was determined from each serum sample using the OIE HI test protocol. 2% and 0.5% cRBC was prepared prior to HA and HI test. Mature chicken was used for the collection of blood for the preparation of red blood cell. For this, sterile syringe and needle containing anticoagulants at the art of 1ml for 9ml blood. Following the collection, it was washed with PBS and centrifuged @1000rpm for 10 min. The suspension of plasma materials and supernatant was discarded and then cRBC was collected. After that 0.5% and 2% cRBC were prepared for slide Haemagglutination test, HA tests and HI test respectively [8]. The unused cRBC suspension was stored at 40C until used. Log<sub>2</sub>X geometric mean titre (GMT) of HI Ab was calculated for each occasion in each group. The data were analyzed with used of paired't' test by Statistical Package for Social Science (SPSS) version 20.0 programme to determine the significant differences in HI titres of pigeon between groups after primary and secondary vaccination [9].

#### **RESULTS AND DISCUSSIONS**

Maternally derived antibody (MDA) of unvaccinated control group B was measured from day 4 to day 49 and the titres were 3.95, 3.05, 1.70, 0.95, 0.30, 0.00 and 0.00 at day 4, 12, 19, 28, 35, 42 and 49 respectively (Figure 2). It was observed that the MDA was persisted at a suitable level up to day 12. But MDA levels started to decline after day 4 and reached at a trace level after 28 day. MDA level became totally zero at day 42. This findings strongly supported by the findings of [10]. He reported that the persistence of MDA up to 27 days of age of birds. [11] was found that the MDA level persisted at a suitable level up to 16 days and it started to decline after 16 days and reached at a negligible level after 26 days. [5,12] who stated that the persistence of MDA in chickens were day 15 to 20 of age. According to [13] HI titre3 (log<sub>2</sub><sup>x</sup>) was protective titre for pigeon. From the result it was also observed that there were no significant differences (p>0.05) of MDA titres between group A and B at day 4. Pigeons of A group vaccinated with a live vaccine Cevac New L® (La Sota strain) at day 5 and day 21 i/o following which sera were obtained from 20 randomly selected pigeons on each occasion of day12, 19, 28, 35, 42 and 49 of age given the HI titre  $(\log_2^x)$  4.75±0.64, 5.65±0.59, 6.85±0.67, 7.55±0.51, 6.35±0.67 and 5.20±0.77 respectively after vaccination (Figure 1). The HI titre increased immediately just after primary vaccination and the HI titre was found



**Figure 1** Comparative HI titre of NDV antibodies in pigeons vaccinated with NDV live vaccines Cevac New L® (La Sota strain) and Control group.

highest after secondary vaccination at day 35. The HI titre was gradually declined after day 35. But this HI titre gave satisfactory protection to the pigeons against ND. After primary vaccination, it was observed that the vaccinated groups of pigeons produced satisfactory immune response compared to those of unvaccinated control group. Intra-ocular vaccination was chosen based on the assumption that this route would make all pigeons received the same dose which was corroborated by reports showing that the ocular route was more efficient [14] and of high antibody titres in white-tailed eagles after intranasal La Sota vaccine [15].

The highest antibody titre was found at day 35 produced by the group A, vaccinated with Cevac New L (Live La Sota strain) and these levels gradually decreased at day 42 and day 49. But HI Ab titer of this group after secondary vaccination increased significantly (p<0.05; day 19 vs. day 35) [16], concluded that secondary vaccination yielded HI titre that was significantly higher than the HI titre after single vaccination. The outcome of the experiment was similar with the findings of [17]. He reported that live lentogenic ND vaccines produced an antibody titre of log<sub>2</sub><sup>4</sup> to log<sub>2</sub><sup>6</sup>. After administration of primary and secondary vaccine A groups responded quickly for the production of Ab and reached highest Abtitre after 2 weeks of vaccination. This was happened because administration of live La Sota strain vaccine which produced higher immune response. But declination process was comparatively slow in case of La Sota strain vaccine. [6] also reported that the best known lentogenic live vaccines were La So ta strain and Hitchner B<sub>1</sub>, both of which have been extensively used in birds. This indicated that La Sota strain of Cevac New L® vaccine might be more immunogenic.

#### **CONCLUSIONS AND RECOMMENDATIONS**

ND is a devastating poultry disease causing about 40%-60% of total annual mortality among the infectious diseases in poultry industry in Bangladesh. Pigeons with infected by ND causing frequently fatal disease primarily associated with neurological signs. Vaccination is the proper way to prevent the disease. In this study, one commercial live vaccine, Cevac New L® (La Sota Strain) was used for the vaccination of the pigeons of group A at day 5 and day 21i/o. It was observed that the mentioned vaccine efficiently induced immune response in pigeons to protect ND.

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The Cevac New  $L^{\otimes}$  (La Sota Strain) vaccine may be used for the prevention and control of ND of pigeons in Bangladesh. Further studies need to develop an appropriate vaccination programme for pigeons with live ND vaccines.

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#### **COMPETING INTEREST**

Authors have declared that no competing interests exist. There is absolutely no conflict of interest between the authors and producers of the products, because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the 1st author.

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