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Original Article

Maternally-Derived Antibody and Seroconversion to Infectious Bronchitis Virus in Chicken

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An investigation was conducted with a view to determining the persistence of maternally-derived antibody (MDA) against infectious bronchitis virus (IBV) in chicks as well as seroconversion of IBV in relation to age and gender of chicks in field condition. Determination of antibody titre was performed by indirect enzyme-linked immunosorbent assay (iELISA). The existence of MDA was calculated in Group A (n = 20) and Group B (n = 15) originating from parents vaccinated against IBV and with no such history respectively. In case of group A, titres of serum samples obtained were 5361.23 ± 854.09, 2567.58 ± 763.61, 808 ± 751.361, 432.29 ± 47.11, 178.36 ± 88.28, 184.58 ± 93.6, 80.89 ± 70.11 and 43.55 ± 32.92 on day 1, 5, 9, 13, 17, 21, 25 and 29 days aged birds respectively. On the other hand, sera samples of group B manifested a titre of 3285.18 ± 685.03, 2219.16 ± 419.77, 1783.62 ± 219.84, 589.01 ± 249.63, 186.66 ± 88.21, 178.36 ± 202.02, 105.77 ± 75.69 and 41.34 ± 69.28 on same schedule. It was further revealed that chicks with ancestor of non-vaccinated parents showed higher MDA titre up to 9th day of age. This might have happened due to exposure of field virus to the parent birds. As regards seroconversion, it was observed that 100, 20 and 80% of serum samples collected from chickens of 0-2 weeks, 3-4 weeks and 4 months of age were positive to IBV specific antibody respectively, whereas no serum sample was positive in case of 5-7 weeks aged birds. When considered the gender of birds, it was revealed that sera from the female showed 58.33% seropositivity against 25% from male. Maternally-derived antibody (MDA) might protect the chickens up to 9 days of age and since MDA could interfere vaccination, it should not be performed before such age.

Keywords: Maternally-derived antibody (MDA), Infectious bronchitis virus (IBV), antibody titre, Seroconversion, Chicks

Introduction

Infectious bronchitis virus (IBV) is a highly contagious pathogen of poultry responsible for very significant morbidity and mortality. The disease is endemic in chicken populations throughout the world[1]. Infectious bronchitis can be controlled by vaccination of chicken flocks; nevertheless, outbreaks still occur in vaccinated flocks due to the lack of cross-protection against antigenically unrelated serotypes and variant strains of the virus[2]. Variant strains of IBV have been recovered from vaccinated flocks despite the use of combinations of several strains of live and attenuated IBV vaccines[3]. Researchers showed that the maternal antibody (MDA) levels might interfere directly with the response to vaccination. The importance of MDA titres in chicks was observed when day-old chicks with MDA showed excellent protection (>95%) against IBV but not at seven days (<30%)[4]. A preliminary study on the prevalence of IBV antibodies in chickens was first reported in Bangladesh by Bhattacharjee *et al.*[5]. To control infectious bronchitis (IB) in Bangladesh, the commercial poultry raisers use IBV vaccine imported from abroad. The reason behind vaccine failure could not readily be established. However, the fact adduced might be (1) serotypic variation of IBV field strain, (2) defective IBV vaccine and (3) influence of MDA.

The present study was undertaken with the objectives to detect the MDA titre in chicks originating from vaccinated and non-vaccinated parents and also to study the seroconversion of IBV in relation to age and gender of chickens in selected areas of Bangladesh.

Materials and Methods

Study area

The study was conducted in the some selected areas in Bangladesh. Blood samples of broiler and layer chicken were collected from the farms of beneficiary of Palli Progoti Sahayak Samity (PPSS) at Faridpur, Society Development Committee (SDC), Talma, Faridpur, Bangladesh Extension Education Services (BEES), Chunarughat and Hobigonj Sadar, Centre for Community

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Development Assistance (CCDA), Putizuri, Hobogonj and brought to the Department of Microbiology and Hygiene, Bangladesh Agricultural University (BAU), Mymensingh for further observation.

Infectious bronchitis virus suspected field serum samples

A total of 44 blood samples encompassing of 20 from different age groups and 24 with equal number from either gender were collected from different areas. Blood specimens were collected aseptically from the left jugular vein using 3-ml disposable sterile syringes and were kept slanted at 4-8°C for overnight to be clotted. Then the clotted blood was removed carefully with sterile needle and serum poured into sterilized graduated centrifuge test tubes. The sera were subjected to centrifugation at 1,000 rpm for 10 min for clarification. Then the clear sera were collected and kept in clean sterilized vials and stored at -20°C for further use.

Serum samples collected from experimental chicks

A total of 48 serum samples with equal number from two groups of experimentally reared chicks were obtained on different interval of age to study persistence of MDA. All chicks were reared up to 29 days. Thus, group A formed with 20 chicks from vaccinated parents stock, while the other group of 15 chicks (group B) originated from parents with no history of vaccination against IBV.

Infectious bronchitis virus-specific antibody detection by iELISA

IBV antibody test kit manufactured by BioChek BV (Crabethstraat 38-C, 2801 AN Gouda, Holland) was used for the estimation of antibody titre of the sera samples of chicks. Briefly, 100 µl of diluted samples were added into the appropriate wells of antigen coated microtitre plate and incubated at room temperature (22-27ºC) for 30 min. Then 100 µl of conjugate reagent was added and again incubated at room temperature (22-27ºC) for 30 min. In next step 100 µl substrate reagent was added and the plate was incubated at room temperature (22-27ºC) for 15 min, finally 100 µl of stop solution was added to stop the reaction. Repeated procedure of washing was carried out where necessary. Appropriate positive and negative control wells were maintained as per direction. The ELISA plate was read by the microtitre plate reader and recorded the absorbance of controls and the test samples by reading at 405 nm. To determine antibody titres to IBV in the test sera, the following calculation was used: anti $\log_{10}$ titre = $1.0^{*}(\log_{10} S/P) + 3.62$. For the antibody status of IBV in chicken, serum samples with S/P value ≥0.2 (titre ≥834), 0.159-0.199 (titre 625-833) and ≤0.149 (titre ≤624) were considered as positive, suspected and negative respectively.

Analysis of data

Data obtained were analyzed by Statistical Package for Social Sciences (SPSS) and Microsoft Excel Programme.

Results

Maternally-derived antibody titre of chicks originated from infectious bronchitis virus (IBV) vaccinated and non-vaccinated parent stock

Sera from Group A at day 1, 5, 9, 13, 17, 21, 25 and 29 of age of chicks originated from IBV vaccinated parent stock showed titre revealed by iELISA were 5361.23 ± 854.09, 2567.58 ± 763.61, 808.85 ± 751.36, 432.29 ± 47.11, 178.36 ± 88.28, 184.58 ± 93.61, 80.89 ± 70.11 and 43.55 ± 32.92 respectively (Figure 1). Here the highest MDA titre was 5361.23 ± 854.09 found in day 1, and the lowest MDA titre was 43.55 ± 3 2.92 found in day 29. In case of Group B, the MDA titre obtained on the respective days were 3285.18 ± 685.03, 2219.16 ± 419.47, 1783.62 ± 219.84, 539.01 ± 249.63, 186.66 ± 88.21, 178.36 ± 202.02, 105.77 ± 75.69 and 41.34 ± 29.28 (Figure 2) in chicks originated from IBV non-vaccinated parent stock; the highest and the lowest MDA titre were 3285.18 ± 685.03 found in day 1 and 41.34 ± 29.28 found in day 29 respectively. However, in both perspectives the MDA titres were positive up to 9 days of age.

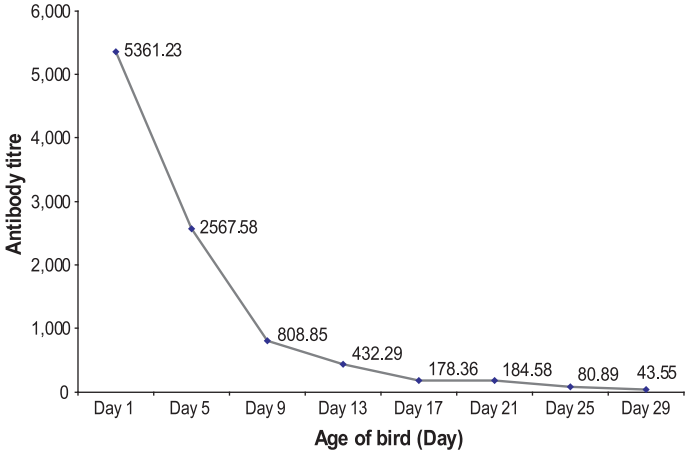

Figure 1. *Line graph of maternally-derived antibody titre of parent vaccinated group of chicken.*

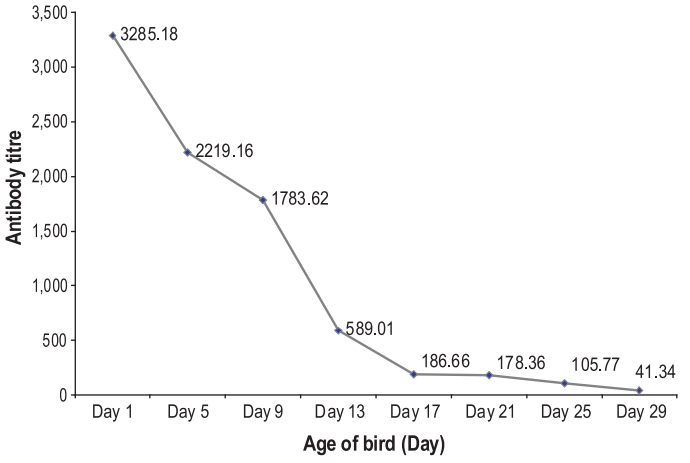

Figure 2. *Line graph of maternally-derived antibody titre of parent nonvaccinated group of chicken*

Seroconversion of infectious bronchitis virus (IBV) on the basis of age

The antibody titre obtained by iELISA of 2 weeks aged birds (sample No. 1-5) were 2651.67, 4000.57, 3589.67, 2786.02 and 1935.11. In case of 3-4-week-old chickens (sample No. 6-10) the

titres were 398.67, 276.75, 840.26, 453.90 and 156.78. In case of the serum samples of 5-7 weeks aged chickens (sample No. 11-15) the titres were calculated to be 43.56, 57.36, 89.56, 56.47 and 90.98. In the last instance, the birds of 4 months of age (sample No. 16-20) exhibited the antibody titre by iELISA were 1033.76, 1265.24, 799.37, 1529.48 and 3672.98. IBV-specific antibody titre reached to 100% positive at 2 weeks of age, 80% positive at 4 month and 20% positive at 3-4 weeks (Figure 3).

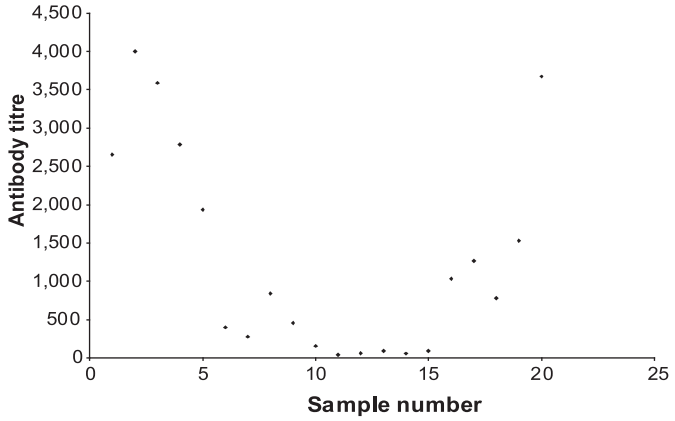

Figure 3. *The horizontal line indicates the positive/negative cut off base line at the titre 834 (on the basis of age).*

Seroconversion of infectious bronchitis virus on the basis of gender

The iELISA antibody titres of female birds (sample No. 1-12) were 1890.87, 3246.34, 682.76, 1009.09, 4675.12, 253.23, 2345.51, 587.07, 2567.01, 146.13, 3987.23 and 267.57. Another 12 male birds showed titres of 2476.15, 345.12, 675.26, 867.17, 574.18, 573.57 and 586.19, 1934.57, 435.89, 299.90, 445.46 and 397.39. It is observed that female birds exhibited higher percentage of positive antibody titre to IBV than male, which was 58% for female and 25% for male. (Figure 4 and 5).

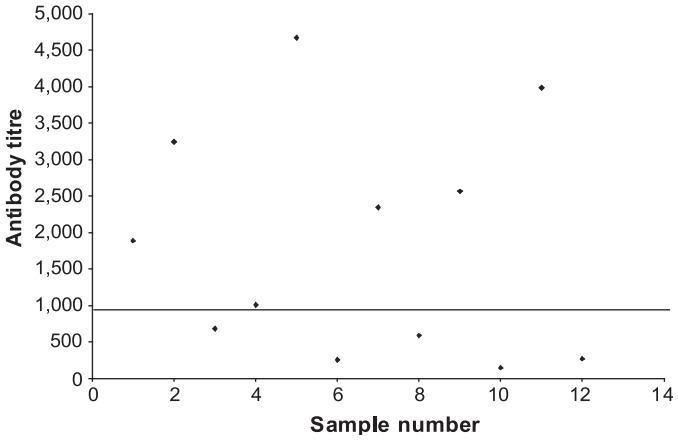

Figure 4. *The horizontal line indicates the positive/negative cut off base line at the titre 834 (female group).*

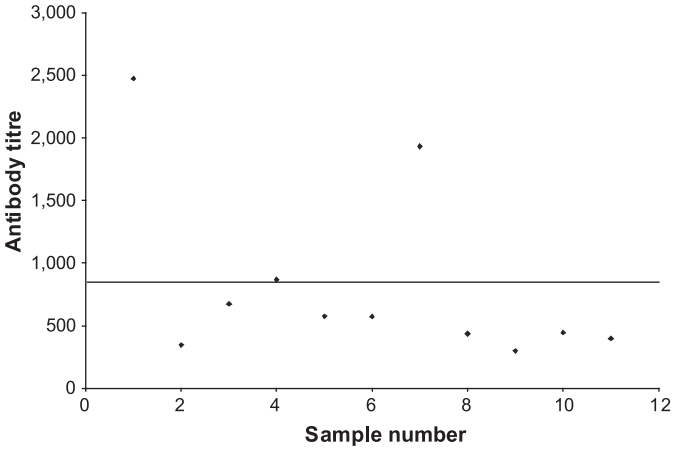

Figure 5. *The horizontal line indicates the positive/negative cut off base line at the titre 834 (Male group).*

Discussion

Outbreaks of infectious bronchitis (IB) still occur, even in vaccinated flocks, and virus strains isolated from those out breaks are often found to be a serotype distinct from the Vaccine type[6]. However, maternally-derived antibody (MDA) can reduce both the severity of vaccinal reaction and the efficacy of the vaccine if the vaccine is of the same type used in the breeder flock immunization[7].

It was observed that MDA titre at first day was 5361.23 ± 854.09 and 3285.18 ± 685.03 in case of chicken from parent vaccinated group and non-vaccinated group respectively. However, the MDA titre was negative in both groups after 9 days of age and remained so until day 29 when the experiment was terminated. Mockett *et al*.[8] showed that MDA provided protection against challenge at day 1 and 1st week, but not at 2nd weeks of age. Chicken from non-vaccinated parent were found positive to IBV antibody at day 1 and decreased to zero around 15 days of age in the experiment of Cardoso *et al*.[9]. Timing of initial immunization might vary due to titre of MDA in chicks and vaccination methods used[9].

From the study on seroconversion, it was observed that IBV-specific antibody was present positively 100% at 0-2 weeks of age. Again 80 and 20% serum samples were positive to IBV specific antibody at 4 months and 3-4 weeks of age respectively, whereas no serum sample was positive to IBV-specific antibody in case of 5 to 7 weeks of age. From such a result, it can be stated that chicks carried MDA up to 2 weeks of age and subsequently it decreased up to 7 weeks. But during 4 months of age, just prior to laying, antibody titre was positive (up to 80%), which justify that just prior to laying chickens are susceptible to IBV. Bhattacharjee *et al*.[5] recorded highest incidence of IB in birds of peak production age group through positive immunofluorescence.

As regards to gender issue, it was observed that female birds showed 58.33% seropositivity, whereas male showed 25% seropositive to IBV. This result is similar to Barbour *et al*.[10] where the authors found that female poultry breeders maintained

antibody titres to IBV up to the 55 days following vaccination, contrary to the males which declined significantly over the same period of time.

References

1. King DJ & Cavanagh D. 1991. Infectious bronchitis. In *Diseases of Poultry* (Calnek BW, Johnbarne H, Beard CW & Yoder HW eds), 9th edn, pp 471-484. Iowa State University Press, Ames, Iowa.
2. Jia W, Karaka K & Parrish CR. 1995. A novel variant of avian infectious bronchitis virus resulting from recombination among three different strains. *Arch Virol*. **140**: 259-271.
3. Gelb JJ, Wolff JB & Moran CA. 1991. Variant serotypes of infectious bronchitis virus isolated from commercial layer and broiler chickens. *Avian Dis*. **35**: 82-87.
4. Mondal SP & Naqi SA. 2001. Maternal antibody to infectious bronchitis virus: Its role in protection against infection and development of active immunity to vaccine. *Vet Immunol Immunopathol*. **79**: 31-40.
5. Bhattacharjee PS, Majumder JU, Miah AH, Ali MI & Rahman MM. 1996. Prevalence of infectious bronchitis antibodies in chickens in Bangladesh. *Bangladesh Vet J*. **30**: 1-6.
6. Cavanagh D & Nyaqi SA. 1997. Infectious bronchitis. In *Diseases of Poultry* (Calnek BW, Barnes HJ, Beard CW, McDougald LR & Saif YM eds), 10th edn, pp 510-526. Iowa State University Press, Ames, Iowa.
7. Klieve AV & Cumming RB. 1988. Infectious bronchitis: Safety and protection in chickens with maternal antibody. *Austr Vet J*. **65**: 396-397.
8. Mockett APA, Cook JKA & Huggins MB. 1987. Maternally-derived antibody to infectious bronchitis virus: Its detection in chick trachea and serum and its role in protection. *Avian Pathol*. **16**: 407-416.
9. Cardoso WM, Gomes LP, Romão JM, Salles RPR, Teixeira RSC, Sobral MHNR, Câmara SR & Oliveira WF. 2006. Antibodies specific to infectious bronchitis in broilers in Ceará state, Brazil. *Arq Bras Med Vet Zootec*. **58**(3): 327-332.
10. Barbour EK, Hamadeh SK, Hilan C & Abbas SS. 1995**.** Comparison of immunity and resistance to diseases in male and female poultry breeders in Lebanon. *Trop Anim Health Product*. **27**(2): 65-70.