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Short Communication

Evaluation of maternal antibody levels for establishing the vaccination program against Newcastle disease in ostrich chicks

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ABSTRACT

Newcastle disease virus (NDV) is known as one of the most important endemic viral pathogen for various avian species such as ostrich, in Iran. Therefore, establishing a routine vaccination program against ND in ostrich flocks would be useful in order to reduce the danger of this infection. Newcastle disease occurs among the ostriches and leads to high rate of mortality while most of the losses are among the youngest ones. This experiment was designed to follow up the changes of maternal antibody in ostrich chicks during the first weeks of their life. At this point of view, 700 one day old ostrich chicks were monitored and every seven days interval 10 blood samples were taken regularly and the titers of maternal antibody in their sera were studied. The haemagglutination inhibition (HI) test was used to evaluate the amount of anti-ND antibody. After hatching this study followed up to 49th day. Due to our findings, the day 30 is recommended as a proper time to start the vaccination program against ND in flocks of ostrich chicks with maternal antibody.

Keywords: Maternal antibody, Newcastle disease, Ostrich

INTRODUCTION

Newcastle disease (ND) is caused by avian paramyxovirus serotype 1 (APMV-1) viruses, which have been placed with viruses of other eight APMV serotypes, in the genus *Rubulavirus*, subfamily *Paramyxovirinae*, family *Paramixoviridae*, order *Mononegavirales* in the current taxonomy (Moro de Sousa *et al* 2000, Rima *et al* 1995). Newcastle disease virus (NDV) occurs worldwide and has a considerable economic impact on the world poultry industry, ranging from losses due to disease and the significant expense for vaccination. Newcastle disease is a serious and important economic disease in ostriches (Moro de Sousa *et al* 2000). Velogenic ND viruses are able to cause disease with high mortality in un-vaccinated ostriches. The disease occurs more often among young ostriches up to one year of age but virulent viruses may affect also adult birds. Samberg *et al* (1989) reported death of 13 of 46 ostriches aged 5 to 9 months, during the outbreak of ND in an ostrich farm. Shanawany (1999)

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noticed that, in ostriches ND might lead to 80 percent mortality in unvaccinated flocks. The hemagglutination-inhibition (HI) test is still the most widely used conventional serological method for measuring anti-NDV antibody levels in poultry sera, and it is considered the standard laboratory test for this disease. However, sera from other species tend to give a high incidence of false-positive results. In order to decrease the false results, it is necessary to either use red blood cells from the same species as the serum samples or to pre-treatment of sera with chicken red blood cells (Alexander 2000). Commercial rearing of the ostrich (*Struthio Camelus*) has been introduced in Iran for at least 10 years. At present, ostriches are kept on many farms, all over the country. Ostriches like many other species of bird are susceptible to Newcastle virus infection. Infection with Velogenic viruses may result in pre acute death in ostrich chicks. As there has not been established an effective treatment for ND in avian, vaccination is still the best way to protect ostriches. Experimental challenge has shown ND poultry vaccines to be protective in ostriches (In higher doses than recommended for chickens). The main objective of the present study is to recommend a proper vaccination program against ND in ostrich chicks with maternal antibody.

MATERIALS AND METHODS

Birds of experiment. Seven hundred ostrich chicks were produced from ostrich farm of Golbarg-e tuba. Three months before hatching the chicks the adult ostriches of the farm had history of vaccination against ND. Ostrich chicks were bled by medial metatarsal vein on days 7, 14, 21, 28, 35, 42 and 49 of the experiment. The blood samples were not taken in day 1, as the chicks are kept in hatchery room until 5th day of their life. Therefore we just notice the mean titer of NDV antibody in their parents that were ≥ 9.4 on that day. Ten blood samples were taken on each turn. Sera were separated and stored at -20C° until serology was performed.

Control group. Control sera were obtained from more than one hundred unimmunized ostrich chicks which were produced from the farms of Markazi province with no history of vaccination in their parents. These ostrich chicks with no maternal antibody were also bled by medial metatarsal vein on days 7, 14 and 56 of the experiment. Titers of 1:8 or greater on day 7 were regarded as positive for previous exposure of parents to NDV and that titers on days 14 and 56 were regarded as positive for exposure of chicks to NDV during the experiment.

Serological method. The HI test was carried out, using 8 hemagglutination (HA) units. This was carried out according to the method described by Stephan *et al* (1998). In our experiment HA test and HI test were performed by using 1% ostrich RBCs; the sera were tested without pretreatment.

Virus antigen. An inactivated virus used as antigen in HI tests which was produced in Razi institute.

RESULTS AND DISCUSSION

Seven days after hatching, significant concentration of antibodies against NDV were detected by HI test in the group of ostrich chicks with the history of ND vaccination in their parents (Table1). During the period of the study the flock was monitored and no clinical signs of ND were reported. The serological findings in control group also confirmed that there has been no challenge with virulent virus (Table2). The minimum titer at the end of the first week (day 7) in the group with maternal antibody was 7 (based on log2) in HI test. At the end of the second week (day 14) the titers were decreased significantly (mean titer 6.9) compared with the titers of the group on day 7

(mean titer 8.8), however all of the samples (10/10) showed titers more than 4(log2) in HI test at this time.

Table 1. Changes of HI titers for ND antibodies in ostrich chicks with maternal antibody in a period of 49 days after hatching.

Dilution	Titer (log2)	Day 7	Day 14	Day 21	Day 28	Day 35	Day 42	Day 49
1/4096	*12*	·	·	·	·	·	·	·
1/2048	*11*	·	·	·	·	·	·	·
1/1024	*10*	**1**	·	·	·	·	·	·
1/512	*9*	**7**	·	·	·	·	·	·
1/256	*8*	**1**	**4**	·	·	·	·	·
1/128	*7*	**1**	**2**	**3**	·	**1**	·	·
1/64	*6*	·	**3**	**4**	**3**	·	**1**	·
1/32	*5*	·	**1**	**2**	**1**	**1**	**1**	**1**
1/16	*4*	·	·	**1**	**6**	**5**	**3**	**4**
1/8	*3*	·	·	·	·	**3**	**3**	**2**
1/4	*2*	·	·	·	·	·	**2**	**2**
1/2	*1*	·	·	·	·	·	·	**1**
No. of Sera		10	10	10	10	10	10	10
Average Titer (log2)		8.8	6.9	5.9	4.7	4.1	3.6	3.2
SD		0.8	1	1	0.9	1.2	1.3	1.2
%CV		9.1	14.5	17	19.2	29.3	36.1	37.5

Table 2: HI titers for ND antibodies in ostrich chicks of control group without maternal immunity.

Dilution	Titer (log2)	Day 7	Day 14	Day 56
1/256	*8*	·	·	·
1/128	*7*	·	·	·
1/64	*6*	·	·	·
1/32	*5*	·	·	·
1/16	*4*	·	·	·
1/8	*3*	·	·	·
1/4	*2*	·	·	·
1/2	*1*	**1**	·	**2**
1/1	*0*	**4**	**5**	**3**
NO. of Sera		5	5	5
Average Titer (log2)		0.2	0.0	0.4
SD		0.4	0.0	0.5
%CV		200	-	125

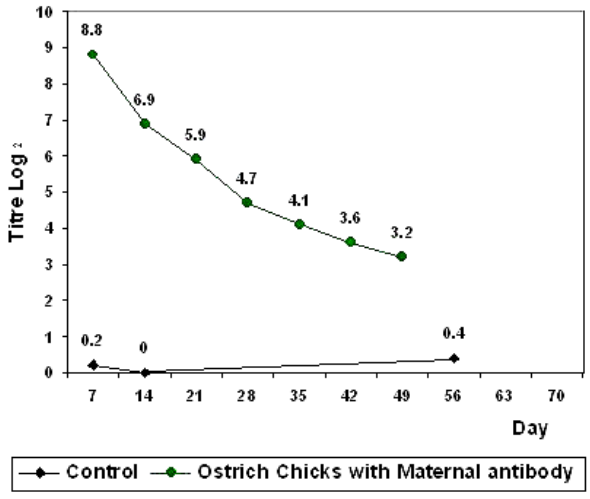

Figure 1. Changes of Mean titer of NDV antibody in ostrich chicks with maternal antibody and with no maternal antibody during the 56 Days after hatch.

On day 21 one out of ten ostrich chicks that were tested showed titer of 4 (log2) in HI test. At the end of the fourth week (day 28) the number of samples in that titer 4 (log2) increased up to 60 percent of the group (6/10). On day 35 of the study, 30% of ostrich chicks (3/10) showed titers less than 4(log2) and 50% of the group (5/10) also were showing titers of 4 (log2) in HI test. A week later (day 42) 50% of ostrich chicks (5/10) showed titers less than 4(log2), 30% (3/10) showed titers of 4(log2), and just 20% (2/10) were showing titers more than 4(log2). At the end of seventh week of the study (day 49), the mean titer was decreased to its minimum level equal to 3.2 (log2). At this time 90% of the ostrich chicks would be at risk, as they did not show titers more than 4(log2) in HI test. Changes of mean titer in both groups of study, (ostrich chicks with and without maternal antibodies) have been shown on Figure 1. Experiments in chickens have shown that HI test titers of ND antibody more than 4(based on log2) are protective against mortality by virulent virus (Nagy *et al* 1991). Cadman *et al* (1997) have accepted this as an agreeable limit for protective ND antibodies titer in ostriches. If we also rely on that presumption, then the following results would be expected; 21 days after hatching, 10% of the ostrich chicks were at border line, although this could be ignored however seven days

later on day 28, the majority of the group as 60 percent of the chicks (6/10) showed titer of 4(log2) in HI test. On day 35, 30% (3/10) of chicks were at risk due to the fact that they showed the titer of 3(log2) in HI test and 50% (5/10) of the group were at the border line (titer of 4). On days 42 and 49 a great number of ostrich chicks were at high risk if they would expose to virulent NDV. Fifty percent (5/10) of these two final groups showed titers less than 4(log2) and even 20% and 30% of them showed titers less than 3(log2) respectively. Madeiros (1997) recommended day 45 would be appropriate for starting vaccination against ND in ostriches. But it is believed that it is too late for starting the vaccination program against ND. Cadman *et al* 1997 noticed that vaccination of ostriches with an oil-based NDV vaccine will lead to reaction producing high levels of antibody in less than eleven days. We also found a good respond using oil-based NDV vaccine in adult ostriches fourteen days after vaccination. On that study the first sampling was established on day 14 after vaccination, therefore changes of antibody before that time was not clear (Momayez 2006, unpublished data).

The study of mean titer changes in the period of 49 days after hatching (Figure1) demonstrates the maternal ND antibodies in ostrich chicks have a half life as about 7 days. However it is not a strict period of time as a half life for maternal ND antibody in ostrich chicks and they lose higher amount of antibody in first 14 days of life compared to the next 35 days of the study. This finding would be useful in order to program a proper time for the first vaccination date. Regarding to the required time for the immune system responding of ostriches after vaccination and also serological findings of this study, we suggest the day 30 after hatching as a proper time for starting the vaccination program against ND in ostrich chicks with maternal antibody.

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