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Serological and Clinical Survey of New Castle Disease in Broiler Flocks of Dezful City, Iran.

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ABSTRACT

The aim of this research to investigate the clinical signs of Newcastle disease (ND) in infected broiler farms in Southwest of Iran and to determine serological status of this flocks compare with healthy flocks. Also the mortality rate was compared in this flock. Serum samples from 9 broiler flocks were collected and examined with Haemagglutination Inhibition (HI) test. Our results showed that, mean of antibody titers in infected flocks was 2.02±0.16 and in healthy flocks was 5.34±0.21.The data was demonstrated that there were significant differences between groups. The mortality rate in infected flocks was 31.35±2.21and in healthy flocks was 10.62±1.15 percent ($p<0.05$). Because of economical losses that caused by growth decrease in ND disease, it is necessary to applying exact and timely vaccination programs in broiler flocks and observe of squeamishly biosecurity to decrease mortality rate and losses due to decrease of growth in this disease.

Keywords: Broiler flocks, Newcastle disease, Haemagglutination Inhibition

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INTRODUCTION

Newcastle disease (ND)is caused by specified viruses of the *AvianParamyxovirus* serotype 1 (APMV-1),which classifies as member of the genus *Avulavirus*, belonging to the family *Paramyxoviridae* [1-3].This highly contagious viral disease, affects domestic poultry and wild birds and characterized by respiratory, gastrointestinal and central nervous system lesions [1,4].

Newcastle disease virus (NDV) causes a disease that varies in clinical severity and transmissibility depending on the pathotype involved. NDV strains are grouped into five pathotypes based on the clinical signs induced in infectedchickens: viscerotropic or neurotropic velogenic with high mortality and intestinal lesions or central-nervoussigns; mesogenic with low mortality, respiratory and nervous signs; lentogenic with clinical mild or inapparent infections of the respiratory tract and asymptomatic enteritic with inapparent intestinal infections [5]. Several factors such as environmental stress, concurrent infections, age of birds and native immunity could affect the outcome of infection. Preventing the virus entering into the field, separating susceptible birds from infected birds, in time vaccination and supportive treatments could be combat the disease [6]. The first outbreaks of ND caused by virulent strains of virus occurred in 1926 in Java, Indonesia and in Newcastle –upon-Tyne, England [7]. Kaleta and Baldauf (1988) concluded that at least 241 species of birds from 27 of the 50 orders of birds are susceptible to NDV. The disease occurs worldwide and has a considerable economic impact on the world poultry industry and it is endemic in poultry industry of Iran and causes economic losses. Also a wide range of avian and non-avian species act as reservoirs of NDV and transmit the disease to susceptible birds [8]. There is long history of NDV recovered from wildlife [9,10]. In Iran, also reported NDV recovered from wildlife [11,12] and domestic chickens [13-16], ostriches [17] and Japanese quail [18]. Recently different diagnostic techniques such as reverse transcription polymerase chain reaction (RT-PCR)proposed by Office International des Epizooties (OIE), that are applied in many laboratories of the world as the most reliable methods for the detection and identification of NDV strains [19]. Considerable populations of industrial chicken farms exist in southwest of Iran especially Dezful city (Khouzestan province), and there was not any report so far published on the economical losses associated with NDV mortality in the industrial broiler flocks in this area of Iran.

This study was carried out to detect the NDV infection, using Haemagglutination Inhibition (HI) serological test and to compare mortality rate between infected and healthy broiler chicken farms of Dezful**.**

MATERIAL AND METHODS

Samples preparations

Totally 135 Blood samples were collected from 9 understudy broiler farms throughout the Dezful city. 1-2 ml blood were collected from brachial vein, allowed to clot at room

temperature then transferred to the laboratory in minimum time and centrifuged at 2500 rpm for 15 minutes, then the sera were collected, labeled and stored at -20ºC for further analysis.

Serological procedure

The serum samples were tested to determine the antibodies against NDV, using the HI test was performed according to the Manual of standards for diagnostic Tests and Vaccination [19]. After making two fold serial dilution of serum up to 11th well, 8 HA unit of NDV antigen (Commercial NDV La Sota Antigen, RAZI Vaccine and Serum Research Institute) was added upto 11th well and kept at 30^{o}C for 30-45 minutes. A 1% chicken RBCs suspension was added into each well. The samples showing peculiar central button shaped settling of RBCs were recorded as positive and the maximum dilution of each sample causing HI was considered as the end point. The HI titer of each serum sample was expressed as reciprocal of the serum dilution.

Statistical analysis

Data were examined using a commercially available statistical package (SPSS version 17 for Windows), and for data analyzing independent samples t-test statistical method was used for compare infected and healthy flocks.

RESULTS

The results indicated that the 4 flocks of 9 were infected with Newcastle disease and 5 flocks were negative. In infected flocks mortality rate was 31.35±2.21 and in non-infected flocks it was 10.62±1.15 ($p<0.05$).

Table1: Mortality rate and HI titer in infected and non-infected flocks

Infected Flocks			Non-Infected Flocks		
Flock No.	Mortality Rate (%)	HI Titre (Log 2)	Flock No.	Mortality Rate (%)	HI Titre (Log 2)
Dez-1-I	23.4	2.60	Dez-1-N	9.1	5.10
Dez-2-I	32.3	1.60	Dez-2-N	12.3	6.30
Dez-3-I	28.5	2.40	Dez-3-N	10.4	6.70
Dez-4-I	41.2	1.50	Dez-4-N	9.7	5.50
-	-	-	Dez-5-N	11.6	3.10
Mean±SE	31.35±2.21	2.02±0.16	Mean±SE	10.62±1.15	5.34±0.21

The HI titer in infected flocks was 2.02±0.16 and in non-infected vaccinated flocks it was 5.34±0.21. The results of HI serological test indicated that the increase of antibody titers in infected flocks was lower significantly than non-infected flocks ($p<0.01$). In non-infected flocks HI titers that was obtained from vaccination was normal and sero-conversion was not seen. In infected flocks greenish diarrhea, depression, reluctant to move was seen and in autopsy green content of gizzard, lesions in intestine was seen, while in healthy flocks there was not any clinical signs or gross lesions.

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DISCUSSION

The HI test is still the most widely used assay that requires cheap reagents, easy interpretation and it is a conventional serological method for measuring anti-NDV antibody levels in poultry sera and considered the standard laboratory method for diagnosis of NDV [5].

Several studies indicated that the respiratory diseases in poultry frequently due to infections caused by numerous factors [20,21]. Prevalence of NDV in broiler chickens is one of the main causes of respiratory diseases and economic losses. ND is endemic in some countries, therefore is as a limiting factor in the development of poultry industrial production in spite of costs for permanent controlling programs against this disease [5,22-24]. Studies in several parts of the world have revealed comparable, lower or higher prevalence of anti-NDV, 72% in Nigeria [25] 43.6% in Ethiopia [26], 28.4% in Viet Nam [27] and 39.1% in Brazil [28]. But in Jordan, researchers were reported that 41.7% of investigated flocks was infected with NDV, of which 13% of flocks infected only with NDV, while in other cases concurrent infections with infectious bronchitis virus (IBV), avian influenza virus (AIV) and Mycoplasma gallisepticum (Mg) [29]. In Iran though most farmers uses vaccines and observe of biosecurity for prevention of ND, a severe form of infection occurred and causing high mortality rate and reduces growth of poultries. Fathi studied 13 flocks of broiler chickens suspected of ND in Chahar Mahal and Bakhtiari province and Rahimian studied this infection in 37 similar flocks in Isfahan province in central area of iran and were reported that 84.6% and 100% of the samples had anti-NDV antibodies [12]. The present paper results as well showed that the 44.5% of understudy broiler farms were infected by NDV. Implicitly, studies have shown that malnutrition, unfavorable weather conditions, levels of maternal antibodies, the challenge virus in the farm, breed quality, quality of vaccines and its administration was effective on ND outcomes [5].

Researchers indicated that the vaccination could not prevent disease occurrence in farm conditions and the findings of them are consistent with the results of this paper. Musa and colleagues studied infection in two flocks of broiler chickens in Nigeria, were reported 100% mortality despite vaccination [24]. Other researchers reported up to 66% mortality during 2002 outbreak of ND in vaccinated flocks of California [30]. The results of the present study, indicated despite the vaccination program, mortality was 23.4 to 41.2 percent, (average: 31.35±2.21) in broiler chicken farms and it could be because of low levels of protection against the velogenic strains of ND, and this results were in consistent with previous studies [24, 30].

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