# Full Length Research Paper

# Evaluation of newcastle disease antibody level after different vaccination regimes in three districts of Amhara Region, Northwestern Ethiopia

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Evaluation of the Newcastle Disease (NCD) antibody level after different vaccination regime was conducted on chickens distributed at the age of day-old in three districts of Amahara region using sera analysis. Three vaccination regimes on chicks against Newcastle using live lentogenic stains; Hitchener B1 (at the age of day-old) and La Sota (at the age of 21st and 60th days of age) were used. The overall antibody level of Newcastle disease in vaccinated chickens using Hemagglutination inhibition test ( $\geq 1.8$ ) was 71.1% ranging from 80.4, 73.3 and 63.3% in Dangila, Bahir Dar and Farta districts respectively. Significant difference (p < 0.001, df = 2,  $\chi^2$  = 13.3) in antibody level of Newcastle disease was found among the three districts. The antibody level of Newcastle disease in Lohmann White breeds (70.6%) was lower than that of Red Island Red breeds (72.5%) but no significant difference was detected between breeds (p > 0.05, df = 1,  $\chi^2$  = 0.27). On the other hand, chickens vaccinated three times at 1st, 21st and 60th day of age (78.8%) were found to have higher protective antibody titer than chickens vaccinated twice at 1st and 21st day of age (64.9%) and only at 1st day of age (65.4%). There was significant difference (p < 0.05, df = 2,  $\chi^2$  = 8.2) in antibody level of Newcastle disease among different vaccination regime and frequencies. In conclusion, the results of the present study indicated that the protective antibody titer response was produced from these vaccines after booster vaccinations. Hence, it is very crucial to vaccinate chickens with the full dose of vaccines against NCD in order to keep protected poultry population.

Key words: Antibody level, chickens, Ethiopia, newcastle disease, vaccination response.

## INTRODUCTION

Newcastle disease (NCD) is among the major constraint to production of village chickens in many developing countries (Sprabrow, 1988; Alexander, 2001). It is the most important viral disease recognized in tropical countries in village poultry production systems. The disease causes great losses in most scavenger and commercial flocks (Sprabrow, 1988; Alders, 2001). Recently, the highly infectious ND is reported to have almost reached 100% mortality in some African countries of the United Republic of Tanzania, Ethiopia and Gambia (Kitalyi, 1997; Tadelle and Ogle, 2001; Tadelle and Jobre, 2004).

NCD is mentioned as one of the disease problems in farms and backyard chickens in most parts of Ethiopia. It has many different local names in different areas and the most common one is "Fengile" (Nasser, 1998; Ashenafi, 2000; Tadelle and Ogle, 2001), which means sudden dorsal prostration and signifies the acuteness and severity of the disease. It is possible to say that currently there are no low risk areas for NCD remaining in Ethiopia. The disease has already become endemic in village poultry population and thus it recurs every year inflicting heavy losses (Tadelle and Jobre, 2004). Vaccination is the most important method of disease control which results in a quite significant increase in chick survival (Spradbrow, 1995; Al-Garib, 2003). Hence, Bureau of Agriculture and Rural Development of Amhara region started a Newcastle disease vaccination program which

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was launched starting from 2003 on day-old chicks distributed to farmers in different districts. Therefore, objective of this paper is to evaluate the antibody level of NCD vaccination on chickens distributed to the farmers.

#### **MATERIALS AND METHODS**

#### Study areas

The present study was conducted in three districts of Amhara region, northwestern Ethiopia. The districts were Bahir Dar Zuria, Dangila and Farta which were considered model districts for poultry development strategy of Amahara Region through distribution of day-old chicks to farmers engaged in day-old chicks rearing practices.

#### Animals and their management

The chicks were obtained from two parents, which were composed of two breeds: Red Island Red (RIR) and Lohmann White (LOH) of replacement parent flocks. These replacement chickens for parent flocks were kept in separate rearing houses. The distance between these houses ranges from 15 - 30 m. There are attendants to provide food and water to the flocks, clean the equipments and collect eggs daily from laying houses and store in cooling houses. The birds were provided with formulated ration obtained from one of the government owned animal feed industries (Kality). The flocks were fed daily twice and given tap water ad lib. They were vaccinated against NCD three times (at 1<sup>st</sup> day old with Hitchener B1 vaccine and at the age of 21 and 60 days with LaSota vaccine).

The chicks were vaccinated against Newcastle disease in Andassa Government Poultry Farm when they are day-old. Two successive vaccinations were undertaken with LaSota strain vaccine by veterinary professionals in the three districts. The day-old chicks were distributed to the farmers with formulated feed enough for two months.

# Study designs

The total day-old chicks distributed to Farta, Bahir Dar Zuria and Dangila districts were 1800, 920 and 910 respectively. Accordingly, with accuracy of 10%, the sample size for Farta district was 180 chicks and 90 chickens each for Bahir Dar and Dangila were considered in this study (Cannon and Roe, 1982; Young et al., 2002). Hence, a total of 360 chicken sera were collected from chickens raised by farmers engaged in day-old chick rearing practices in the three districts. Information were collected on chicken owners and veterinary professionals selected as study units from each district to obtain relevant data on the number and age of vaccinated chickens, the strain of vaccine used and date of distribution of day-old chicks.

#### Serum collection

About 100  $\mu$ l blood was collected from the wing vein chicks above 90 days of age with non-heparinzed vacutainer tube of 5 ml and 23 G (32 mm). The tube containing the blood was kept at room temperature overnight in slanting position until the blood clot and the serum was transferred into a sterile eppendorf tube. The tubes were labeled and stored at -20 °C until analysis.

#### Hemagglutination-Inhibition (HAI)

HAI test was done according to the procedures indicated in of OIE (2004). The test was conducted in the National Veterinary Institute (NVI), Debre zeit-Ethiopia. The test was carried out by running two fold dilutions of equal volumes (25 µl) of Phosphate Buffered Saline (PBS) and test serum (25 µI) in U-bottomed micro titer plates. 4 Hemagglutination units of (HAU) the viral antigen of LaSota strain obtained from CIRAD, France was added to each well and the plates were left at room temperature for a minimum of 30 min. Finally 25 µl of 1% (v/v) chicken RBCs collected from four Specific Pathogen Free chickens older than 3 weeks and serologically negative to NCD antibody was added to each well and, after gentle mixing, the plates were allowed to settle for about 40 minutes at room temperature. The HI titer was read from the highest dilution of serum causing complete inhibition of 4 HAU of antigen. Those wells that showed sedimentation of RBC as the control wells (containing only 25 µl RBCs and 25 µl PBS) were considered as inhibition. A titer greater than or equal to 1:8 was taken as positive.

#### Statistical analysis

The data were analyzed using descriptive statistics and Chi-square test for comparison of antibody levels across the districts, breeds and vaccination frequencies.

### **RESULTS**

The overall antibody titre of Newcastle disease in vaccinated chicks ( $\geq$ 1:8) was 71.1% ranging from 80.4, 73.3 and 63.3% in Dangila, Bahir Dar and Farta districts respectively. Significant difference (p < 0.001, df = 2,  $\chi^2$  = 13.3) in antibody level of Newcastle disease was found among the three districts. The NCD vaccination antibody titer in LOH breeds (70.6%) was lower than that of RIR breeds (72.5%) and no significant difference was detected between breeds (p > 0.50, df = 1,  $\chi^2$  = 0.27) (Table 1).

On the other hand, chickens vaccinated three times at  $1^{st}$ ,  $21^{st}$  and  $60^{th}$  day of age (78.8%) were found to have higher protective antibody titer than that of chickens vaccinated twice at  $1^{st}$  and  $21^{st}$  day of age (64.9%)) and only at  $1^{st}$  day of age (65.4%). There was significant difference (p < 0.05, df = 2,  $\chi^2$  = 8.2) in antibody level of Newcastle disease among different Newcastle vaccination frequencies (Table 2).

# DISCUSSION

The overall population with protective antibody titre (≥1:8) (71.1%) in the study districts, which was above 70%, indicated that vaccinated day-old chicks have protective immunity against Newcastle disease. This is in line with the epidemic theory which suggests that if 70% of the population is immune, the disease outbreak is unlikely to occur because there are not enough susceptible to propagate an epidemic (Thrusfield, 1995; Young et al., 2001).

Table 1. Antibody titer of Newcastle disease in vaccinated chickens of two breeds across three districts.

Antibody titer	Breed		District			
	RIR	LOH	Bahir Dar	Dangila	Farta	Overall
	(n = 91)	(n = 269)	(n = 90)	(n = 90)	(n = 180)	(n = 360)
Positive (≥1:8)	66 (72.5%)	190 (70.6%)	66 (73.3%)	76 (84.4%)	114 (63.3%)	256 (71.1%)
Negative (<1:8)	25 (27.5%)	79 (29.4%)	24 (26.7%)	14 (15.6%)	66 (36.7%)	104 (28.9%)

Table 2. Newcastle disease antibody titer level across different vaccination regimes.

	Frequency of Vaccination					
Antibody titer	1 <sup>st</sup> day (n = 52)	1 <sup>st</sup> & 21 <sup>st</sup> day (n = 148)	1 <sup>st</sup> , 21 <sup>st</sup> & 60 <sup>th</sup> day (n = 160)	Overall (n = 360)		
Positive (≥1:8)	34 (65.4%)	96 (64.9%)	126 (78.8%)	256 (71.1%)		
Negative (<1:8)	18 (34.6%)	52 (35.1%)	34 (21.2%)	104 (28.9%)		

The higher population with protective antibody titre in Dangila (84.4%) and Bahir Dar (73.3%) districts compared to Farta (63.3%) district was attributed to the accomplishment of more booster NCD vaccination frequencies in the two districts than Farta district. Moreover, the population with protective antibody titre was higher in those chicks which were vaccinated three times (78.8%) than those vaccinated two times (64.9%) and only once (65.4%). The highest number of protected population in chicks vaccinated three times is due to the fact that booster dose vaccination of NCD was applied on chickens (Spradbrow, 1995; Alexander, 2001; Al-Garib, 2003).

Based on the results of this NCD antibody level response, the author is convinced that vaccination of davold chicks against NCD using the vaccine of Hitchener B1 (at the age of day-old) and LaSota strain vaccines (at the age of 21st and 60th days of age) produced from National Veterinary Institute, Debre zeit-Ethiopia formed protective antibody level and it is very important to vaccinate chickens against NCD in order to keep protected population against NCD virus infection. Moreover, the present study indicated that the chicks distributed in Bahir Dar and Dangila districts were found to be protected. Higher protective antibody level was detected in chickens vaccinated three times than those vaccinated twice and only once indicates that booster vaccination of chickens against Newcastle disease vaccine created significant protective antibody titre.

In conclusion, it is paramount important that the Newcastle Disease vaccination strategy should be redesigned and continued to apply both on commercial and village chickens to decrease the loss associated with mortality of chickens caused by Newcastle disease.

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