# Different Vaccination Programs against Newcastle Disease in Broiler Chickens

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### **ABSTRACT**

One hundred fifty four broiler chickens were divided into 6 groups, 14 chickens in group 1 and 28 chickens of each in groups 2 to 6. Groups 1 to 4 received different Newcastle disease (ND) vaccination programs, these were: group 1 received live ND C2 vaccine at 1-day-old; group 2 received live ND C2 vaccine at 1-day-old and a booster vaccination with live ND Clone 30 (NDC30) vaccine at 10-day-old; group 3 received live ND C2 simultaneously with inactivated oil adjuvant ND (IOAND) vaccines at 1-day-old and group 4 received live ND C2 simultaneously with IOAND vaccines at 1-day-old and a booster vaccination with NDC30 vaccine at 10day-old. Both live ND vaccines were given by coarse spray and inactivated vaccine was given by subcutaneously injection at the base of the skull. Groups 5 and 6 did not receive any vaccine and served as positive and negative controls, respectively. All chickens of groups 1 to 5 were challenged at 35-day-old. Mortality was recorded for 10 days after challenge. Chickens were bled at 10-, 28, 35 and 45-day-old. Sera were collected and tested for antibody titres against ND virus by haemagglutination-inhibition (HI) test. The results revealed that mortality among the vaccinated chickens after challenge were 42.86, 17.86, 10.71 and 0 percent for groups 1, 2, 3 and 4, respectively. All challenge and non-vaccinated chickens died, indicating that there was no resistance of this non-vaccinated control group of chickens. Antibody titres of vaccinated birds were also corresponding to mortality rate. It is concluded that the simultaneous vaccination with live ND C2 and IOAND vaccines at 1-day-old and a booster vaccination with NDC30 vaccine at 10-day-old gave 100 percent protection against ND virus challenge.

Key words: Newcastle disease, Vaccination, Antibodies, Protection, Broiler chickens

## INTRODUCTION

The basic principle for Newcastle disease (ND) prevention is the same as other diseases, these are, good management practice and biosecurity or good hygiene in conjunction with vaccination. It has been reported (Bennejean *et. al.*, 1978) that simultaneous vaccination of day-old chicks with live and killed ND vaccines results in

better protection as compared with a single vaccination. However, our previous study found that simultaneous vaccination of day-old chicks with live and killed ND vaccines had only  $88.57\pm9.00$  percent of survival chickens after chickens were challenged at 28-day-old (Chansiripornchai and Sasipreeyajan, 2006). In the present study, the addition of a booster vaccination at 10-day-old was performed and compared to other vaccination programs. Protection was determined 10 days after chickens were challenged with viscerotropic velogenic ND virus (vvNDV) at 35-day-old.

#### MATERIALS AND METHODS

#### CHICKENS

One hundred fifty four female Abor Acres broiler chickens were divided into 6 groups, 14 chickens in group 1 and 28 chickens of each in groups 2 to 6. Birds were raised in cages and each group of bird was kept in separate room but similar environmental conditions. Feed and water were provided *ad libitum*. Chickens were weighed at 10, 35 and 45-day-old. Guidelines and legislative regulations on the use of animals for scientific purposes of Chulalongkorn University, Bangkok, Thailand were followed.

#### VACCINES AND VACCINES' ADMINISTRATION

Two live ND vaccines; ND C2 (Nobilis® ND C2, Intervet international B.V., Boxmeer, Holland.) and ND Clone 30 (NDC30) (Nobilis® Ma5+Clone 30, Intervet international B.V., Boxmeer, Holland.) were given by coarse spray at 1-day-old with the droplet size of 235 micrometres ( $\mu$ ) and 10-day-old with the droplet size of 173  $\mu$  (Desvac, France), respectively. According to the manufacturers' recommendations, each vaccine is in 1000 dose vials, one dose being at least 105.5 and 106 50% embryo infective dose (EID50)/bird, respectively. Inactivated oil adjuvant ND (IOAND) vaccine (Nobilis® ND BROILER, Intervet international B.V., Boxmeer, Holland.) was given 0.1 ml (50PD50/bird) by subcutaneously injection at the base of the skull at 1-day-old.

#### VACCINATION PROGRAMS

Chickens of groups 1 to 4 received different ND vaccination programs, these were: group 1 received ND C2 vaccine at 1-day-old; group 2 received ND C2 vaccine at 1-day-old and a booster vaccination with NDC30 vaccine at 10-day-old; group 3 received ND C2 simultaneously with IOAND vaccines at 1-day-old and group 4 received ND C2 simultaneously with IOAND vaccines at 1-day-old and a booster vaccination with NDC30 at 10-day-old. Groups 5 and 6 did not receive any vaccine and served as positive and negative controls, respectively.

#### CHALLENGE STUDY

Chickens of groups 1 to 5 were challenged with vvNDV strain (ICPI=1.8) by oral drop at 35-day-old, at a dose of 106 EID50/chicken. Mortality was observed for 10 days after challenge. Dead chickens were necropsied and gross lesions were determined.

## SEROLOGICAL EXAMINATION

Chickens were bled at 10, 28, 35 and 45-day-old. Sera were collected and tested or NDV antibody titres by the haemaglutination-inhibition (HI) test as described by Van Eck and Goren (1991).

## STATISTICAL ANALYSIS

Body weight and antibody titres were analyzed and compared between groups using ANOVA and Duncan's multiple range test with SPSS 9.0 software. Mortality was calculated by using Chi-square values. Differences between groups were considered significant at p<0.05.

## RESULTS AND DISCUSSION

There were significant difference of body weight of chickens at 10-day-old (p<0.05) but at the time of challenge at 35-day-old, they were not significant difference (p>0.05) (Table 1). During the period of 10 to 35-day-old, there were 2 chickens died. One chicken in group 4, there was due to vaccinal reaction. Another chicken in group 6, there was due to ascites. Vaccination reaction is quite common after live vaccine administration, especially by spray (Van Eck, 1990). However, in this experiment, there was no noticeable vaccinal reaction after live ND C2 vaccination which is probably due to 2 reasons, these are, interference between vaccine virus and maternal antibodies and the low pathogenic nature of the vaccine virus. Some degrees of reaction were observed after a booster vaccination with live NDC30 at 10-day-old. There was no detrimental effect due to vaccinal reaction except one chicken of group 4 died. It was reported that clone 30 vaccines giving by spray had less vaccinal reaction than uncloned La Sota vaccines (Al-Garib, et al., 2003).

Antibodies at 10-day-old were maternal antibodies which were in between  $2.77\pm0.87$  to  $3.00\pm0.98$  and were not significant dfference (p>0.05). Antibodies at 28 and 35-day-old compared among the vaccinated groups, group 4 which received simultaneous ND C2 and IOAND vaccines and a booster vaccination with NDC30 at 10-day-old, had the highest titre corresponds to the highest protection (Tables 2 and 3). Titres at the day of challenge at 35-day-old was  $4.26\pm1.63$  which gave 100 percent protection. However, this level of titre is lower than mean level of 5.2 which was presented by Allan *et al.*, (1978) as 100 percent protection. Our earlier work (Chansiripornchai and Sasipreeyajan, 2006), in which the chickens received simultaneous live and IOAND vaccines at day-old an received the challenge virus at 28-day-old, had  $88.57\pm9.00$  percent protection which is similar to the protection result of group 3 of the present study (Table 3).

Chickens of group 1 had the same low level of antibody titres as non-vaccinated control but the protection was 57.14 percent. It is probably due to local immune response in the respiratory tract (Al-Garib, *et al.*, 2003) which was not measured in this study. While non-vaccinated chickens had no protection. Dead chickens were necropsied and gross lesions were observed. The lesions confirmed ND virus infection depicted by conjunctivitis, tracheitis, airsacculitis, pneumonitis and haemorrhage on proventriculus, small intestine, caecal tonsil, rectum, heart muscle and coronary fat. At the end of the experiment at 45-day-old, the survival chickens had higher antibody titres due to anamnestic response. There were no titre change in non-vaccinated and non-challenge chickens but their body weight were among the highest.

#### **CONCLUSION**

It is concluded that chickens received live ND C2 at 1-day-old, live ND C2 at 1-day-old and a booster vaccination with live NDC30 at 10-day-old, live ND C2 simultaneously with IOAND vaccines at 1-day-old, live ND C2 simultaneously with IOAND vaccines at 1-day-old and a booster vaccination with live NDC30 at 10-day-old gave 57.24, 82.14, 89.29 and 100 percent protection, respectively. While non-vaccinated and challenge control chickens had no protection.

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Table 1. Body weight of chickens at 10-, 35- and 45-day-old.

Group	Body weight (mean $\pm$ SD, gm/bird)				
	10-day-old	35-day-old	45-day-old		
1	$291.43 \pm 24.21^{\circ} (14)^{A}$	$1,464.29 \pm 72.19^{a}$ (14)	$1,707.50 \pm 171.78^{a}$ (8)		
2	$237.14 \pm 20.48^{b}$ (28)	$1,480.71 \pm 59.87^{a}$ (28)	$1,760.00 \pm 142.09^{a}$ (23)		
3	$239.46 \pm 18.07^{b}$ (28)	$1,465.71 \pm 136.72^{a}$ (28)	$1,833.60 \pm 169.75^{a,c}$ (25)		
4	$247.86 \pm 12.87^{b}$ (28)	$1,472.59 \pm 107.62^{a}$ (27)	$1,761.11 \pm 187.95^{a}$ (27)		
5	$214.64 \pm 18.46^{a}$ (28)	$1,530.00 \pm 134.25^{a}$ (28)	_ B		
6	$212.86 \pm 20.16^{a} (28)$	$1,483.33 \pm 81.48^{a}$ (27)	$1,904.44 \pm 268.71^{\circ}$ (27)		

A Number of chickens in the group

 $<sup>^{</sup>a,b,c}$  The different superscript in each column means statistically significant difference (p < 0.05)

B All chickens died

Table 2. Serological response before and after NDV challenge.

Group	NDV antibody titers ( $\overline{x} \pm SD$ ; log <sub>2</sub> )				
	10-day-old	28-day-old	35-day-old	45-day-old	
1	2 05 + 0 00 a (22)A	$1.00 \pm 0^{\circ} (14)$	$1.00 \pm 0^{\circ} (14)$	$7.88 \pm 4.39^{a}$ (8)	
2	$2.95 \pm 0.90^{\text{ a}} (22)^{\text{A}}$	$3.73 \pm 3.04^{a,b}$ (22)	$2.96 \pm 2.15^{a}$ (28)	$5.48 \pm 3.70^{a} (23)$	
3	$3.00 \pm 0.98^{a}$ (22)	$2.73 \pm 1.49^{b}$ (22)	$3.25 \pm 1.51^{a}$ (28)	$10.40 \pm 4.56^{\text{ b}}(25)$	
4	$2.91 \pm 1.02^{a}$ (22)	$4.23 \pm 2.18^{a}$ (22)	$4.26 \pm 1.63^{b} (27)$	$7.30 \pm 3.86^{a} (27)$	
5	$2.91 \pm 1.07^{a}$ (22)	$1.00 \pm 0^{\circ} (22)$	$1.00 \pm 0^{\circ} (28)$	- <sup>B</sup>	
6	$2.77 \pm 0.87^{a}$ (22)	$1.00 \pm 0^{\circ} (22)$	$1.00 \pm 0^{\circ} (27)$	$1.00 \pm 0^{\circ} (27)$	

**Table 3.** Mortality rate of chickens after challenge.

Group	Mortality rate		Precent
	Number A	Percent	Protection
1	6/14 <sup>c</sup>	42.86	57.14
2	5/28 b 3/28 b,d	17.86	82.14
3	3/28 b,d	10.71	89.29
4	0/27 <sup>d</sup>	0	100
5	28/28 a	100	0
6	0/27 <sup>d</sup>	0	100

 $<sup>^{\</sup>rm A}$  Number of serum samples tested  $^{\rm a,b,c}$  The different superscript in each column means statistically significant difference (p < 0.05)  $^{\rm B}$  All chickens died

<sup>&</sup>lt;sup>A</sup> Number of dead chickens/total chickens in the group a,b,c,d The different superscript in each column means statistically significant difference (p < 0.05)