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## Field trial of Malaysian thermostable Newcastle disease vaccine in village chickens in Kaduna State, Nigeria

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### **Abstract**

Village chickens in Kaduna State, Nigeria were vaccinated once with a Malaysian heat-resistant Newcastle disease vaccine (NDV4HR) given in feed. In all, 1605 chickens in 223 households covering 33 villages and 13 Local Government Areas were tagged and bled before vaccination and two weeks after vaccination. Antibodies to Newcastle disease virus were titrated by haemagglutination inhibition test and titres  $\geq 3(\log_2)$  were assumed to be protective. Presumed protective titres

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were recorded in 143 (8.9%) of chickens before vaccination and in 957 (65.5%) after vaccination. Recommendation is made for the widespread adoption of this technology.

**Keywords:** Newcastle disease vaccine; Antibody response; Vaccination; Village chicken; Thermostability.

## Introduction

Newcastle disease (ND) is reported as the most important viral disease of poultry in the world including developing countries (Nawathe et. al., 1975; Adu et. al., 1986; Adene, 1990; Spradbrow, 1997). It has a devastating effect on commercial as well as village poultry industries (Philips, 1973; Nawathe et. al., 1988; Okeke and Lamorde, 1988; Shamaki et. al., 1989; Adene, 1997). The resources derivable from the chickens cannot be fully utilized unless the disease is controlled particularly in the village poultry flocks that are believed to keep the virus circulation and act as reservoirs and carriers to themselves and the more susceptible exotic breeds in commercial farms (Nawathe et. al., 1975; Ezeokoli et. al., 1984; Gomwalk et. al., 1985; Adu et. al., 1986; Nwosu and Okeke, 1989; Olabode et. al., 1992).

Vaccination has been reported as the only safeguard against endemic ND (Orajaka et. al., 1999; Usman, 2002). The village chickens, in multi-age flocks scattered in small number over villages, are difficult to catch for formal vaccination as adopted in commercial chicken enterprise (Aini et. al., 1990; Orajaka et. al., 1999; Usman, 2002). It therefore takes great effort and time trying to vaccinate them. More importantly, these conventional vaccines are not heat-labile and therefore require complex cold-chains to link the vaccine producers and users (Aini et. al., 1990; Usman, 2002).

Preliminary work conducted elsewhere indicates that heat-stable a virulent V4 or 1-2 strains selected for heat resistance and applied through eye drop, drinking water or food and feed particles has been found to be a suitable oral vaccine for village chickens (Saglid and Spalatin, 1982; Spradbrow, 1988; Spradbrow, 1992; Bell et. al., 1995; Tu et. al., 1998). This V4 or 1-2 vaccine has been successfully used in many African countries such as The Gambia (Jarra et. al., 1991), Cameroon (Bell et. al., 1995), Ghana (Amakye – Anim et. al., 1998) and many countries of South-East Asia (Copland, 1987). Maize byproducts after processing have been found to be good carriers of V4 in the vaccination of birds against ND in Nigeria (Olabode, 1996).

In Malaysia, a 60% protection rate was recorded on challenge of village chickens with virulent NDV strain and farmers in Malaysia have benefited from the oral food vaccination of their flocks with NDV4HR (Aini et. al., 1990). It therefore became necessary to assess the immunological status of Nigerian village chickens and to conduct field trials with this newly developed vaccine for the purpose of determining its suitability as a rural poultry vaccine in Kaduna State, Nigeria. In view of the economic importance of ND in Nigerian village chickens, indications are that local poultry farmers would welcome ND vaccinations capable of protecting their flocks. The objective of this study was to investigate the seroprevalence antibody status of ND in Nigerian local chickens and to conduct field vaccination trials with ND4HR in village chicken flocks in Kaduna State, Nigeria. The outcome of the results obtained would form the basis for recommendations on whether the new vaccination technology will be adopted in Kaduna State, Nigeria.

## Materials and Methods

### *The study area*

The study covered thirteen (13) Local Government Areas of Kaduna State located within the semi-arid and sub-humid zones of North Central Zone of Nigeria. Kaduna State is situated between 8° 45' and 11° 30' N and 6° 10' and 9° E. The mean annual temperature is about 34°C with the hottest months being from March – April (40°C) and the coldest period (13.2°C) is between December and January during the severe harmattan. Rainfall varies between 1,000mm and 1,500mm and rainy season lasts for 150-200 days (Mid April-end to October). The dry season occurs from late October to early April (RIM, 1993).

### *Flocks Sampling Procedure*

Prior to the survey, group meetings were held with the Management Staff of the State Veterinary Department and the Heads of Agricultural Department of the 23 Local Government Areas (L.G.A.'s) of the State for discussion on the project objective, benefits, sample size and areas to be covered. At the meeting, it was decided that a total of 2,000 village chickens in 250 households and 40 villages be sampled in two local government areas of the State (Kaduna North and South) based on high population density of village chickens in the areas. Out of the list of 60 villages supplied by the State, 40 villages were randomly selected through balloting and out of a total of 400 households listed by the village heads of the 40 villages selected, 250 households were also randomly selected.

Due to poor response from the majority of the farmers who refused to allow their chickens to be bled for fear of death, despite the offer of free poultry drugs and feeds as incentives to them, only 300 village chickens in 50 households and 10 villages were sampled initially. Additional 11 Local Government Areas consisting of 50 villages, 250 households and 2000 village chickens were listed for sampling based on the areas with high concentration of village chickens and presence of United Nations Development Programme (UNDP) activities on agriculture.

Scheduled visits were made to the additional proposed Local Government Areas and villages in the company of the LGAs representatives and meetings were held with the village heads and farmers to discuss the objectives of the project, its benefit and the areas to be sampled. The list of the villages and households were supplied by the village heads. From the list, additional number of 1,305 village chickens in 173 households of 23 villages were selected from the 11 Local Government Areas that responded. Thus a total of 1,605 village chickens in 223 households covering 33 villages and 13 Local Government Areas were selected for the study.

### *NDV4HR vaccine*

The NDV4HR used for the field trials in a freeze-dried live thermostable strain vaccine imported from Malaysian Vaccines and Pharmaceutical SNP, BHD. Each vial of the vaccine contains 100 bird doses. It is a lentogenic live virus selected for its heat resistance. The vaccine dose per bird was 106.0 EID<sub>50</sub> (50% embryo infective doses).

### *Vaccination of Chickens and Blood Sampling*

Vials of the NDV4HR were reconstituted in the feed as recommended by the manufacturer and administered to the birds. The chick mash used was obtained from Sanders Feeds (SEEPN Nigeria Limited), Kaduna Depot, Nigeria. The chick mash contains 19.00% crude protein, 6.00% crude fat, 1.25% calcium, 0.65% phosphorus, 1.00% lysine, 0.80% methionine, 5.00% crude fibre and 2700 kilocalorie per kg metabolisable energy. The required dose per flock in each household was calculated based on the recommendation of 10g of the feed per bird (Aini et. al., 1990). In the calculated amount of feed was added an equivalent amount of diluent (well water), 10ml of well water containing 106.0 EID<sub>50</sub> of vaccine per 10g of feed. The moist mixture was then put in a clean feeding trough under a shade for the birds to consume. All the chickens in the flocks selected were vaccinated. About 50% of the chickens in each vaccinated flock were caught and wing-tagged, bled and released. Two weeks post vaccination, the wing-tagged birds were bled again. A total of 1,605 wing-tagged village chickens were bled before and after vaccination. Both pre and post vaccination sera were tested for NDV haemagglutination inhibition (HI) antibodies.

#### Newcastle Disease Virus Antigen Preparation

The antigen was prepared from NDV-LaSota vaccine obtained from the National Veterinary Research Institute, Kaduna Field Station. The 200 dose vial of the NDV-LaSota vaccine was reconstituted in 8ml distilled water. The haemagglutination (HA) titre was determined as described by Beard (1980) and diluted to contain 4 HA

units for use in the HI test as described by Allan and Gough (1974).

#### Data Analysis

Number of birds with detectable NDV antibody titres were calculated. Sera with HI titres  $\geq 3(\log_2)$  were considered positive or protective based on the reports of Allan et. al., (1974); Westbury et. al., (1984); Aini et. al., (1990), Chi-square was used to determine the relationship between the number of village chickens that had antibody titre  $\geq 3(\log_2)$  pre and post vaccination periods.

#### Results

Out of a total of 1,605 village chickens screened for NDV antibody, 143 (8.90%) had antibody titre  $\geq 3(\log_2)$  before vaccination (Table 1). Also out of the 143 birds that had protective antibody titre  $\geq 3(\log_2)$  prevaccination, only 135 (94.41%) had protective titre  $\geq 3(\log_2)$  post vaccination. Very few proportion (5.59%) of the birds that tested positive before vaccination tested negative after vaccination.

About 65.46% (95% CI = 63.02 % < P < 67.88%) of the birds that had antibody titre  $\leq 3(\log_2)$  before vaccination sero converted to higher HI titre  $\geq 3(\log_2)$  post vaccination (Table 1). Presumed protective titres were recorded in 1092 (68.04%) of birds post vaccination (Table 1). There was a statistically significant difference in the frequency of protective titre pre- and postvaccination ( $X^2 = 48.87$ , 1df,  $P < 0.001$ ).

**Table 1:** Number of birds with protective antibody titres to ND pre and post vaccination

		Post vaccination		
		Titre $\geq 2^3$	Titre $\leq 2^3$	Total
Pre-vaccination	Titre $\geq 2^3$	135 (94.4%)	8 (5.6%)	143
	Titre $\leq 2^3$	957 (65.5%)	505 (44.5)	1462
	Total	1092 (68.0%)	513 (32.0%)	1605

#### Discussion

In this study, the HI titre Titre  $\geq 3(\log_2)$  were considered positive (protective) based on the findings of Allan et. al., (1974); Westbury (1984) and Bell et. al., (1991a) who reported that birds with HI titres Titre  $\geq 3(\log_2)$  were protective against challenge with a virulent strain of ND virus. The protective antibody titres  $\geq 3(\log_2)$  recorded in 143 (8.9%) out of 1605 village chickens screened for HI antibody before vaccination suggested previous exposure of the birds to field strains of ND virus (velogenic). This is in agreement with the report of Alexander (1998) who showed that in all paramyxoviruses, birds that have not been immunized or infected usually have HI titres  $\geq 3(\log_2)$  and that non-specific titres above this levels are rare in avian species. Also farmers and veterinary staff interviewed reported that no ND vaccination has been conducted in these villages and their respective neighbourhoods during the past 12 months prior to the study. Reports of ND outbreaks in the monthly returns from the Local Government Veterinary Officers, Ezeokoli et. al., (1984) and personal communications with the farmers confirmed ND as a widespread problem in the study area. It has also

observed that a small proportion (5.6%) of the birds that were protected prevaccination, tested negative post vaccination. This observation contrasts with the previous findings of Bell et. al., (1991b) who found that positive breeders (naturally exposed) responded serologically and that the number of birds with titres  $\geq 3(\log_2)$  increased significantly following vaccination. In addition, about 65.5% of the birds that had antibody titres  $\geq 3(\log_2)$  before vaccination seroconverted to protective level ( $\geq 3(\log_2)$ ) post vaccination. The statistically significant difference in the frequency of protective titre prevaccination and post vaccination signifies that the vaccine, NDV4HR, is immunogenic and capable of provoking antibody response (Aini et.al., 1990).

#### Conclusion

Recommending the use of NDV4HR to Nigerian rural farmers requires that the local conditions, protective ability of the vaccine and ease of administration is taken into consideration. In Kaduna State of Nigeria, where farming is predominant, many farmers are yet to be convinced of the benefits of ND vaccination. Achieving good level of protection with the use of NDV4HR in vaccination of chicken will stimulate the interest of rural farmers in adopting the new vaccine technology.

The use of NDV4HR in a single oral vaccination of village chickens in this study, has achieved a 65.5%

protection. Therefore, subsequent application of more doses of the vaccine at intervals to be determined by further research may result in the development of a higher protection in greater proportion of chickens and subsequent reduction in the outbreaks of ND. Finally, controlling ND in the endemic rural areas of Nigeria through the use of NDV4HR will subsequently control the spread of the ND virus in the neighbouring countries such as Cameroon, Tchad, Niger and Benin Republic.

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