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Quality Assessment of the Efficacies of Some Commercially Used Newcastle Disease Vaccines in Jos, Plateau State, Nigeria.

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Abstract

The efficacies of some of the commercially used Newcastle disease vaccines in Jos were determined in fifty (50) exotic white leghorn birds with hemagglutionation inhibition (HI) and Egg infective dose 50% (EID_{50}) tests. By culturing in bacteriological media the vaccines were sterile. Vaccinated birds were safe with no clinical infections; NVRI Lasota and komarov vaccines had post vaccination antibody titre of 512 HI units and 256 HI unit and booster antibody of 2048 HI unit and 1280 HI unit respectively. Post vaccination antibody in ABIC lasota was 192 HI unit, BIOVAC was 128 HI while the booster was 1024 in both ABIC and BIOVAC. NVRI vaccines gave better immune antibody production than ABIC and BIOVAC. Over 80% of the birds survived post vaccination challenge with 10^5 log_{10} LD_{50} Hertz33 challenge ND strain. Although the vaccines protected the birds at different levels, it is advisable to use vaccines with indigenous ND strain for better protection. Other information are discussed.

Key words: Vaccine, Newcastle disease, Antibody, birds, efficacies

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Introduction

Newcastle disease is a fatal contagious viral disease of poultry birds although all birds are infected (Alexander, 1997; Samad, 2005). It is caused by an enveloped, non-segmented, negative sense, single stranded RNA virus belonging to genus Rubulavirus and family paramyxoviridae (Alexander 1998). The disease is characterized by coughing, gasping, sneezing, watery greenish or whitish diarrhea, respiratory distress, swelling of the head and neck, (Olabode and Chukwuedo, 2005; OIE, 2009).

Infection in birds may be asymptomatic, subclinical to very acute disease with high mortality and susceptibility varies from species to species in birds. Newcastle disease is a dreaded disease of poultry with a huge economic loss in infected birds. It is one of the important economic disease causing big set back in the rapidly growing poultry industry in Nigeria (Alexander, 2000; Chukwuedo, 2005; Ibu *et al.,* 2009). An average of 200-250 outbreaks of the disease is reported in Nigeria annually (Okeke and Lamorde, 1988).

The control of ND in poultry is by the use of live potent attenuated Newcastle disease vaccines with good management practices (Van Eck *et al.,* 1990; Chukwuedo and Olabode, 2003; Hassan *et al.,* 2006). However, in the recent times there have been reported cases of post vaccination outbreaks with Newcastle disease in poultry. This may suggest vaccine failure or emergence of new ND virus strain in birds which can not be controlled by the use of the present vaccines. In line with the current problem of vaccine ineffectiveness, this study was designed to determine the efficacies of some commercially sold ND vaccines in Jos and its environs.

Materials and Methods

Vaccines: Three (3) brands of Newcastle disease lasota vaccines and one (1) brand of Komarov vaccine were purchased from commercial Vendor. They are NVRI ND-Lasota ND-Komarov (National Veterinary Research Institute, Vom. Nigeria). ABIC ND-Lasota (ABIC Ltd., Netanya, Israel) and BIOVAC ND-Lasota (BIOVAC Ltd, Akiva, Israel). The vaccines were maintained in cold temperature (20^0C in deep freeze).

Birds: Fifty (50) exotic white leghorn birds were purchase from Access farm in Ibadan, Oyo State, through ECWA Veterinary farms Bukuru in Jos South Local Government of Plateau State. The birds were 3 weeks old and were divided into five (5) groups of ten birds per room with adequate water and feed. Each group was given one type of the vaccines.

Antiserum and antigens: The standard antiserum and antigen were obtained from virology Research division of the National Veterinary Research Institute, Vom. They were tested and stored at -20^0C ready for use.

Serum samples: The birds Pre - and post - vaccination blood serum samples were collected. 5ul of blood was collected per bird. The sera were heat inactivated in a water bath at 56^0C for 30 minutes. They were store at -20^0C ready for use.

Chicken red blood cells: The chicken red blood cells were collected from 6-8 week old bird known to be NDV antibody free via the wings branchial vein into universal bottle containing sodium citrate anticoagulant. The cells were washed three times in PBS, PH 7.2 and the pack cell volume (PCV) determined the red blood cells were store at 4^0C fridge temperature until used.

Hemagglutination test (HA): The HA tests were carried out in U-shaped polystyrene disposable plates with 96 wells. 50ul of PBS was added into wells 2 to 12 and the vaccine antigen suspension was added in 50ul volume in wells 1 and 2. The content of well 2 was serially diluted in 2-fold serial dilution to well 12. 50ul of 1% chick rbc was added to all wells including the controls and the plates were incubated at 4^0C for 45 minutes after which they were read.

Hemagglutination inhibition test (HI): The HI test was done in U-shaped polystyrene disposable plate with 96-wells. 50ul of PBS was added in well 2 through to well 12. The test serum was added in wells 1 and 2. The content of well 2 was serially diluted in 2-fold serial dilution to well 1.50ul of the antigen suspension (4HA unit) of previously titrated antigen was added to the test well and controls. The plates were incubated at 4^0C for 30 minutes to allow antiserum antigen reaction to take place. 50ul of 1% chick red blood cells was added to all wells including controls and the plates well re-incubated at 4^0C for 45 minutes after which the tests were read.

Egg Infective Dose 50% (EID50): The egg infective dose 50% was done by inoculating 0.1ul of the various vaccine dilutions in day old chicken embryonated eggs. 10^{-1} to 10^{-10} dilutions of vaccines were prepared and five eggs were inoculated per dilution. The inoculated eggs were incubated at 37^0C for 72 hours before they chilled at 4^0C over night and their allantoic fluid spot tested with 10% chick rbc. The eggs are scored positive by evidence of hemagglutination.

Sterility test: The vaccines were individually reconstituted with PBS based on the manufacturer's guideline. 100ul of each of the vaccine were inoculated into bacteriological and mycological media. These include blood agar (BA), Nutrient agar (NA), MacConkey agar (MCA), Sabouraud dextrose agar (SDA) and Thioglycholate broth (TGB). The media were incubated at 37^0C for 72hours and observed daily for growth of contaminant (Okeke, 1984).

Safety test: One hundred micro litres (100ul) of the various vaccines were separately inoculated into their respective birds. The birds were observed daily for morbidity or mortality (OIE, 2009)

Potency test: All the 10 birds in each of the groups given the different vaccines separately and the unvaccinated control birds were challenged with 10^{-5} LD_{50} (50% lethal dose) of Hertz 33 (ND challenge virus). Where 90% of the vaccinated birds survivals and all the control birds died within one week (7 days) is considered potent vaccine (FOA, 1984).

Results

Table 1: Sterility and Safety Test results

Vaccine	Media					Safety Rate
Types	BA	NA	MCA	SDA	TGB	(%)
NVRI Lasota	-	-	-	-	-	9/10 (90%)
NVRI Komarov	-	-	-	-	-	9/10 (90%)
Biovac Lasota	-	-	-	-	-	9/10 (90%)
ABIC Lasota	-	-	-	-	-	8/10 (80%)

Key: BA =Blood agar, MCA = MacConkey agar, NA = Nutrient agar, SD =Sabouraud dextrose agar, TGB= Throughlycholate broth, - = Negative for microbes, + = Positive for microbes

Table 2: HA and EID_{50} Titres of the Vaccine

Vaccine	Mean Titres		
	Spot	Titration	EID_{50}
Type	HA	HA	Titration
NVRI LAsota	++	512	$10^{-10.5}$
NVRI know	++	256	$10^{-8.25}$
BIOVAC Lasota	++	256	$10^{-10.5}$
ABIC Lasota	++	512	$10^{-9.5}$

Key: HA = Hemagglutination test, EID_{50} = Egg infective dose 50%,

+ = Week agglutination, ++ = Strong agglutination

Table 3: HI Titres and Post Challenges Survival

	Mean Titres			
Vaccine	Pre Vac	Post Vac	Booster Vac	Post challenge
Type	HI	HI	HI	Survivals (%)
NVRI Lasota	<2	512	2048	90%
NVRI Komarov	<2	256	1280	100%
BIOVAC Lasota	<2	128	1040	100%
Control Birds	<2	<2	<2	0%

Key: HI = Hemagglutination Inhibition test

Results

Vaccination of poultry birds against Newcastle disease (ND) and with good management practices are very important as an effective means of protecting poultry from the serious economic losses of 70-80% mortality in poultry annually caused by ND virus infection (Van Eck *et al.,* 1991; Westbury, 2001; Chukwuedo and Olabode, 2005). At the time of vaccination many farmers do not screen to check the birds ND virus antibodies status before vaccination as well as improper administration of the vaccines may lead to vaccine failure (Chukwuedo and Mbakwe, 2007).Some time the farmers choice of vaccine strains as well as methods of vaccination adopted depends on the cost of vaccine, cost of vaccination, availability of vaccine and the antigenicity of vaccine, virus and virulence of field virus (Van Eck, 1990; Olabode and Chukwuedo, 2005). The prevalence of velogenic strains of Newcastle disease virus in free roaming local birds may serve as reservoir of the virus causing outbreaks which the birds that have been vaccinated with foreign vaccines may not be protected against (Islam *et al.,* 2008, spradbrow, 1992).

In this present study the sterility and safety tests showed that the vaccines are safe in birds with no microbial contaminants that may interfere with the birds post vaccination immune responses. This is in line with OIE (2009) report that no vaccines should contain any adventitious agents(Table 1). The spot HA test gave strong agglutination for all the vaccines with chick rbc and the HA titration titres were above 128HAU required for immunization of the birds. NVRI and ABIC Lasota had 512 HA titres while NVRI Komarov and BIOVAC lasota had 256 HA titres. The EID_{50} infective virus titres were above $10^{-7.5}$ base line immunizing titre in all the tested vaccines. The NVRI and BIOVAC lasota vaccines had $10^{-10.5}$ $\log_{10}$ EID_{50} titres followed by ABIC Lasota with $10^{-9.5}$ $\log_{10}$ EID_{50} while the NVRI Komarov had the least titre with $10^{-8.25}$ $\log_{10}$ EID_{50} (Table 2).

The pre- and post vaccination titres in the birds showed the vaccines were quite immunogenic. The pre – vaccination antibody titre was <2HI unit in all the birds suggesting no maternal ND antibody in the bird. The post vaccination mean antibody was quite above 128 HI unit which is required to protect the birds. NVRI Lasota and Komarov had 512 and 256 HI units respectively. The ABIC had 192 mean HI titre while the BIOVAC had the least with 128 HI unit. The booster vaccination showed evidence of hyper

immune antibody production. NVRI Lasota had the highest antibody (2048 HI unit), the Komarov had 1280 HI unit while the ABIC and BIOVAC had 1024 HI unit (Table 3).

The control of Newcastle disease in endemic region like Nigeria may be very difficult especially with the presence of many ND susceptible scarvenger bird around and the use of foreign vaccines. More often than not, it requires repeated vaccination of the birds which may increase cost and make the farmers not to adhere to full vaccine application regiment (Olabode and Chukwuedo, 2005; Chukwuedo and Mbakwe, 2007). The results of this study may have shown that NVRI vaccines has greater immune responses and advantages in the birds than the foreign vaccines based on the high protective HI antibody. However one bird was lost in NVRI Lasota group after challenge due to rodent attack (Table 3). Meanwhile where there is scarcity of the vaccines any of these vaccines may be used since post vaccination challenges gave over 80% survivals with all the vaccines. It is therefore advised that poultry farmers should strictly adhere to the manufacturer's guideline in the use of Veterinary vaccines for better protection and production. The post vaccine failure or disease outbreaks may be attributed to other factors like poor nutrition, poor management practices, host parasitic infection and inadequate vaccination of the birds and not on the quality of the vaccines.

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