EVALUATION OF THE EFFICACY OF NEWCASTLE DISEASE (LASOTA) LIVE VACCINES SOLD IN JOS, PLATEAU STATE, NIGERIA

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Abstract

Introduction: Newcastle Disease is a highly contagious and commonly fatal viral poultry disease. The study was carried out to determine the efficacy of Newcastle Disease (NDV) vaccines sold at open market in Jos.

Materials and methods: Five commonly used brands of ND LaSota live vaccines were used for the study. Haemagglutination test (HA) and Egg Infective Dose (EID_{50}) was carried out on the vaccines.Sixty (60) day-old unvaccinated white leghorn broilers were distributed into 6 groups n=10 and allowed to acclimatize for 6 days. Groups 1-5 were vaccinated intraoculally with vaccines coded A - E respectively, while group 6 was not vaccinated (control group). They were bled on days 7, 14, 21 and 28 post vaccinations (PV) for antibody titration using standard methods. They were challenged

with a velogenic strain ($10^{5.5}$ CID_{50}) of NDV day 35 and observed for morbidity and mortality for 14 days.

Results: HA test revealed viral titres of $7log_2$-$9log_2$ for vaccines with egg infectivity dose (EID_{50}) of $10^{7.00}$ to $10^{8.49}$. There was progressive increase in antibody titre following vaccination. There were 100% mortalities among the control group and non in test groups. Performance of the indigenous vaccine was commendable.

Conclusion: we therefore, conclude that the vaccines sold at Jos, Plateau State are potent and could prevent NDV infection and they are reliable, if standard protocols are observed. The use of the indigenous vaccine brand should also be encouraged to prevent introduction of a new strain into the country.

Keyword: Antibody, Titre, Broiler Chickens, EID_{50}, Vaccines, NDV.

Introduction

Newcastle disease (ND) is a highly contagious and commonly fatal viral poultry disease affecting mainly domestic and wild avian species (Spradbrow*et al.,* 1990). The disease caused by Newcastle disease virus (NDV); an avian paramyxovirus serotype designated APMV-1. APMV-1 is further classified into three pathotypes based on their virulence in chickens which are: lentogenic, mesogenic and velogenic (OIE 2004 andAlexander, 2003). The disease is characterized by respiratory symptoms such as coughing, gasping, sneezing and rales. Other signs include dropping wings, dragging legs, swelling of tissues around neck and eyes, twisting of the neck, circling and cessation of egg production (Wakamatsu *et al.*,2006). Human infections via exposure to infected birds can cause mild conjunctivitis and influenza-like symptoms and in severe cases, it can lead to some lasting impairment of vision (Beard *et al.,* 1984).

Newcastle disease is worldwide in distribution (Chu and Rizk 1972), causing an epizootic problem in Nigeria (Ezeokoli*et al.,* 1994 and Olabode*et al.,* 1992) and as such having great negative economic impact in the nation. The presence of this virus can also limit trade and the development of intense poultry production resulting to major constraint to the availability of protein for human consumption (Nwankiti*et al.,* 2010).

The disease has no cure; however, the control of the disease relies on the regular use of safe and effective vaccines (Spradbrow 1992). Live vaccines prepared with lentogenic strains of NDV are commonly used for broilers than vaccines prepared from chemically inactivated strains, mixed with adjuvant (Alexander 1997 andBigg*et al.,* 1988). Freeze-dried NDV live vaccines can be produced on a large scale at a relatively low cost. The vaccines are easy to administer on a large scale, and rapidly stimulate

humoral, cell-mediated and mucosal immunity (Chandraseker et al., 1989, Perry and Aitken 1973).

Despite the continuous use of NDV vaccine in poultry confinements for the control of the disease, pocket of infection have been reported in vaccinated flocks (Roy et al., 2000). Vaccine failure may be responsible for such outbreak. It has caused many farmers to lose confidence in locally produced vaccines, whereby, avoiding the indigenous brand questioning its efficacy. The poultry farmers prefer to vaccinate their birds with imported vaccines. It therefore, became necessary to verify the potency and efficacy of the locally produced vaccines, comparing with foreign brands and give appropriate recommendation to the relevant agency.

Materials and Methods

Sample source and size:

Five different brands of Newcastle disease LaSota Live (NDV-LL) Vaccines sold at the metropolitan market of Jos, Plateau State of Nigeria were obtained from major veterinary stores. The brands include HIPRA, IZOVAC, INDOVAC, ABIC and NVRI represented as A, B, C, D and E. The vaccines obtained and transferred under cold chain and standard bio-safety practice to the virology laboratory, NVRI Vom. One of these vaccine brands is an Indigenous brand (NVRI), others from Spain,Isreal, Italy, India etc.

Day old chickens and Specific pathogen free eggs

Sixty(60)unvaccinated day-old leghorns broiler chickens where obtained from the animal livestock department, National Veterinary Research Institute, Vom, Plateau State of Nigeria and housed in the experimental animal house of the institute. 9days old embryonated specific pathogen free eggs were also obtained from hatchery section of this research institute. The study location is situated at the northern middle belt of Nigeria; Longitude $8^{o}37'30''$and latitude 9^{o} 45'50'.

Egg infective dose at 50% (EID_{50}) and haemagglutination (HA) test.

The EID_{50} of the various brands of vaccine were determined using the technique by Reed and Muench1938). HA tithe were determined by treating a serially diluted 50µl of each vial suspension with 50µl of 0.5% of chicken red blood cells in saline and incubated at room temperature ($28\pm1^{o}C$) according to method described by Allan and Gough (1974).

Treatment

During the entire study the broilers were fed with feeds carefully compounded to meet 23% crude protein (CP) and 3200Kcal. Metabolizable

energy (ME) for broilers, starter and 20% CP and 3000Kcal for broiler finisher, it was ensured that the level of mycotoxins in feed were maintained relatively low throughout the experiment, the quality and quantity of groundnut cake (GNC), soya bean and rice bran included in the feeds were determined by biurette method (Ranja 1999) while the Metabolizable energy was determined by the Bomb Calorimeter method (AOAC 1980). The broilers were housed in battery cages of 0.31 m^2/ bird as recommended by Mustafa et al. (2010). All experimental protocols complied with NIH guidelines(National Research Council, 1985) as approved by the ethical and research committee, NVRI, Vom. They all received all necessary medications, allowed to acclimatize for 6days, fed with standard feeds and water *ad libitum.* The birds where distributed to six groups (A, B, C D, E and F) n=10 caged separately. Group F is the control group. All groups except Group F were administered a corresponding vaccineintraocularly (0.003ml dropper), and booster dose intramuscularly with 0.1ml of NDV komoroffon day 21. All Groups were challenged intra muscularly with 0.1ml of a velogenic field strain of NDV ($10^{5.5}$ chicken infective dose) on the 35^{th} day post vaccination. The birds were observed for morbidity and mortality for another 14days.

Antibody assay

Venous blood was harvested from the wing veins on day 1, 7, 14, 21 and 28 post vaccinations, allowed to clot and serum separated using bench centrifuge at 1500r. p. m for 10 min. They were assay to determine HI titre for NDV antibody according to protocol described in OIE, Terrestrial Manual (2004). However, the birds were bled before vaccination on day 1as pre vaccination titre.

Statistical analysis

The data collated from the study in various treatment groups were statistically analyzed using one way analysis of variance (ANOVA). Significant differences between the treatment means were determined at 99% confidence level.

Result

The HA titre, EID_{50} and percentage mortality of the tested vaccines are shown on table 1. The HA titre of the vaccines were adequate, ranging from $7log_2$ to $9log_2$. The EID_{50} of the various brands of vaccines were also higher than the $10^{6.5}$/dose minimum virus level for ND LL vaccines. The mortality rate following viral challenge was 100% in control group and there was no loss in groups vaccinated with Vaccines A, B, C, D and E.

The pre and post vaccination antibody titre (log_2) of broiler chickens are shown on table 2. The pre vaccination antibody titre (baseline) of the broilers was insignificant ($<2log_2$) in all the groups. On day 14 post vaccination, the antibody was observed to be increased showing significant titre ($>4log_2$) in all the groups except group D (ABIC) which recorded $3.8log_2$. On day 28, the titre was higher in A (HIPRA) and E (NVRI) and least in D (ABIC) which recorded $7.3log_2$. The titre of the control group was not significant ($<4log_2$) throughout the period of study. There was however, no statistically significant difference in the antibody titre produced by the various vaccinated groups at $P = 0.01$ on the various days.

Discussion

The safety of vaccines should be carefully monitored, starting early in the production development and continuing for as long as the vaccine is being used. Pharmacovigilance should be encouraged to determine the adverse events following vaccination of birds. It has been earlier documented that vaccines preventable infection have decreased and the spotlight of public health media concern has shifted to vaccine safety (Hofacre 1986).

The result of the current study yielded suggestion that there is no justification to continue to rely on the imported ND LL vaccine, since the locally produced one is reliable. It also suggests the discontinuous reliance on imported NDV vaccine because of likely variations in field strains, if the storage chain is broken unnoticed or unintentionally; it may lead to revert of genomics and may introduce a new strain of NDV to the community.

Instead of distancing themselves from the use of locally produced vaccine because of the perceived failure or pocket of adverse reactions. Healthcare providers should report without delay all serious or unexpected adverse events following vaccination to the public health officer or veterinarian who will in turn transmit same to the government agency handling vaccine pharmacovigilance in Nigeria. However, the event (either seriously or adverse) following vaccination may be a side effect of the vaccine, since all drug/ vaccine have side effects.

The immunity observed in vaccinated birds indicated that the vaccines were potent. The antibody titre built-up in the tested birds was significant and certified the minimum viral level for ND LL vaccines. The two fold increase in humoral responses of the birds following ‘primer dose’ and ‘booster dose’ was commeasurable with the efficacy of the imported vaccines and the corresponding HAtitre. This is in tandem with the work of Abbas *et al.* (2006), Spardbrow*et al.* (1988) and Alexander (2003), who in their various report documented on NDV dose–response relationship among the virus content, serological response and clinical protection. It is also in agreement with the report of Muhammed*et al*. (2006) and Chandrasekar*et al.*

(1989), who in separate documentations, stated that there is unusual gradual increase in ND antibody titre following the primer vaccination and a higher response to a secondary booster dose.

The mean HItitre was $6.1\log_2$ ($6.5\log_2 – 5.6\log_2$) on day 21, it was significant and sufficiently high to confer immunity against NDV on the birds, thereby protecting them from adverse effect of the challenge strain. This finding is also in tandem with the work of Allan *et al.* (26) who reported a mean HI titre of $5.2\log_2$ as adequate against NDV infection. The mortality rate was 100% in unvaccinated birds because they were not protected either with primary or secondary dose of the vaccine. The significant effect of vaccines could be explained by the principle of active immunization against infectious diseases in the management and control of viral infection in poultry birds. The clinical signs following challenge of both vaccinated and unvaccinated groups further substantiate the effect of vaccines on the management of poultry diseases. This report is not at variance with the earlier report of Ojiezeh*et al.* (2013) who highlighted the relevance of vaccination in disease prevention, control and management and stated that vaccine produced locally are potent against NDV disease.

Among the five vaccines tested, HIPRA and NVRI performance was higher followed by IZOVAC and INDOVAC. Though, ABIC was a performing vaccine but with a lower antibody titre of 5.60 ± 0.54 on day 21 and 7.30 ± 0.21 on day 28. This devalue may not be unconnected to poor handling of the vaccine on storage.

Conclusion

The vaccine produced locally is potent and efficient against NDV infections in poultry. Farmer should be encouraged to patronize the locally produced vaccines and report adverse event following vaccination to stakeholders in pharmacovigilance group especially when cold chain-biosafety protocol have not been compromised. The performance of locally produced NDV vaccine is commendable.

Recommendation

- Continuous monitoring of the safety of vaccines sold in open markets in Nigeria.
- Identify increases in the frequency and severity of previously identified vaccine-related reactions.
- Identify previously unknown adverse event following immunization that could possibly be related to a vaccine.
- Identify areas that require further investigation and/or research.

- Provide timely information on adverse events following vaccination reporting profiles for vaccines marketed in Nigeria, that can help inform immunization-related decisions.
- Formation of vaccine vigilance working groups whose responsibility would include:
    - Preparation of national guidelines and procedures for monitoring and management of adverse effect following vaccines.
    - Provide national form to identify, share and promote best practices regarding vaccine pharmacovigilance and
    - Provide a national vaccine safety sentinel network that can rapidly share and disseminate information to appropriate stakeholders regarding vaccine safety issues and signals.

Conflict of interests

The authors do not have a direct financial relationship with any of the commercial identity mentioned in this paper.

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Table 1:Haemagglutination (HA) titre, Egg infective dose at 50% (EID50), and Mortality percentage

Vaccine	HA titre (log_2)	EID_{50} (log_{10})	Percentage Mortality (%)
HIPRA	8	8.5	0
IZOVAC	7	7.7	0
INDOVAC	7	7.4	0
ABIC	9	7.0	0
NVRI	8	8.7	0
CONTROL	Nil	Nil	100

Table 2: Antibody titre post vaccination (log_2) of broiler chickens

Group	***Day 0***	***Day 7***	***Day 14***	***Day 21***	***Day 28***
A	<2	2.30 ± 0.48	4.30 ± 0.68	6.40 ± 0.52	8.00 ± 0.15
B	<2	2.60 ± 0.52	4.10 ± 0.32	6.10 ± 0.94	7.60 ± 0.27
C	<2	2.50 ± 0.71	4.00 ± 0.67	5.70 ± 0.48	7.40 ± 0.22
D	<2	2.10 ± 0.57	3.80 ± 0.42	5.60 ± 0.54	7.30 ± 0.21
E	<2	2.80 ± 0.42	4.50 ± 0.71	6.50 ± 0.97	8.10 ± 0.23
F	<2	<2	<2	<2	<2

- $<4log_2$ is insignificant titre.
- $>4log_2$ is significant titre.
- All values are means ± SEM of 10 birds per group.