

## Seroprofile of Antibodies to Fowl Poxvirus in Commercial and Indigenous Chickens in Southwestern Nigeria

<sup>1</sup>O.G. Ohore, <sup>1</sup>B.O. Emikpe, <sup>3</sup>D.O. Oluwayelu, <sup>1</sup>R.O. Adeyemi and <sup>2</sup>M.A. Ockiya

<sup>1</sup>Department of Veterinary Pathology,

<sup>2</sup>Department of Medicine and Veterinary,

<sup>3</sup>Department of Microbiology and Parasitology, Faculty of Veterinary Medicine,  
 University of Ibadan, Ibadan, Nigeria

**Abstract:** This study was carried out to determine the sero-prevalence of Fowl Poxvirus (FPV) antibodies in both local and exotic poultry in some states of south western, Nigeria using the Enzyme-Linked Immunosorbent Assay (ELISA) technique. A total of 552 serum samples from farms in 4 states of southwestern Nigeria, Ogun, Ondo, Oyo and Lagos states as well as 184 sera of indigenous chicken from various households were obtained for the study. Of this, 248 samples from 3 farms were from vaccinated flocks while 304 samples from 3 other farms were from non-vaccinated flocks against Fowl Pox (FP). An overall prevalence of 80% was obtained for the non-vaccinated chickens. Of this, the local chicken showed 89% prevalence, growers, 10% layers, 75 and 80% in breeders, a prevalence of 95 -97% in layers and 100% was observed in layers and breeders, respectively in the vaccinated flocks. Within the states where samples were collected, 80% prevalence was observed in Lagos state and 75% in Oyo state. There were no significant differences between the prevalences in the groups except for the grower type that was significantly lower than the others. The mean  $\pm$  standard deviation of the positive sera was higher in local chicken (1.350+134) when compared to all the other groups including the vaccinated birds ( $p < 0.001$ ). There was no significant difference ( $p < 0.05$ ) between the titres obtained in the vaccinated layers and breeders and between the non vaccinated layers and breeders. The vaccinated breeders, however, had significantly higher mean titres ( $p < 0.005$ ) than the non-vaccinated breeders. The result showed that fowl pox is endemic in both exotic and indigenous poultry in southwestern Nigeria. The results also showed that there was a significantly higher response in the local breeds to FPV infection than in the exotic breeds, as has been observed with other disease agent.

**Key words:** Seroprofile, antibodies, fowl poxvirus, commercial and indigenous chickens, Nigeria

### INTRODUCTION

Nigerian poultry production consists of both exotic and indigenous breeds. The industry is of immense importance especially as source of protein and it provides employment for youth and women. Adene (1997) reported that indigenous poultry account for over 70% of Nigerian poultry; hence they are very important in the epizootiology of poultry diseases. Indigenous birds are usually kept on free-range management system and not normally vaccinated (Gueye, 1998). They are characterized by survival traits such as small body size, slow growth, late maturity and small production ability (Ibe, 1990).

The exotic breeds of poultry are usually reared in intensive systems of management either in battery cages or in deep litter system and usually perform sub-optimally in the tropics compared to the local chickens which are more adapted to such tropical conditions as high temperature and humidity (Mark *et al.*, 1969). It is believed

that the free wandering local chickens act as potential reservoirs and carriers of infection to themselves and the more susceptible exotic breeds in commercial enterprises (Adene *et al.*, 1985; Adu *et al.*, 1986).

Fowl pox caused by Fowl Poxvirus (FPV) is a slow spreading disease characterized by the development of discrete nodular proliferative skin lesions on the non-feathered parts of the body (cutaneous form) and proliferative lesions on the upper respiratory tract, mouth and oesophagus (diphtheritic form) (Tripathy and Reed, 2003). It usually present with high morbidity and mortality. Although vaccines are available, farmers do not routinely use them and recently, they have been field reports of post vaccination outbreaks.

Sero-epidemiological surveys of some diseases of poultry in Nigeria had been done such as Marek's disease (Adene, 1983) infectious bursal disease (Adedokun and Durojaiye, 1999) Newcastle disease (Ohore *et al.*, 2003) egg drop syndrome 76 (Adedokun and Durojaiye, 1999)

and Fowl typhoid (Ohore *et al.*, 2002). There is however, very little information on the epizootiology of fowl pox especially in indigenous chickens.

In this study, the emphasis is to redirect focus on fowl pox with a view to monitoring its serological activity in the poultry industry using the Enzyme-Linked Immunosorbent Assay (ELISA) technique.

## MATERIALS AND METHODS

**Test samples:** The test samples for the study were sera obtained from indigenous and commercial chickens (growers, layers and breeders) from four states in southwestern Nigeria including Lagos, Ogun, Ondo and Oyo states. A total of 552 samples consisting of 368 commercial poultry and 184 indigenous chicken serum samples, were selected randomly from various flocks in 6 farms and several households, respectively in south-western Nigeria for the indirect Enzyme-Linked Immunosorbent Assay (ELISA) for fowl poxvirus antibodies.

### Bird management

**Indigenous chicken:** The indigenous (local) chickens were raised under the extensive or semi intensive systems of management in which only shelter and sometimes kitchen wastes are provide as feed to the birds, which are mainly scavengers.

**Commercial poultry:** The exotic commercial poultry were raised intensively in properly managed pens and kept either in battery-cages or deep-litter system.

**Selection of chickens:** Representative samples were obtained from different states (Oyo, Ogun, Ondo and Lagos) in southwestern Nigeria (Fig. 1). For each farm where samples were collected, the vaccination history especially with regards to fowl pox was obtained from their records. All chickens sampled were apparently healthy as at period of sampling. Neither age nor breed was used as a criterion for selection. Indigenous chickens were sampled from Oyo state only due to logistic considerations.

**Chequerboard titrations for determination of optimal dilution of analytes:** The optimum dilution for each of the analytes (antigen, serum and conjugate) in the test procedure was determined empirically by a chequer board titration. The antigen used was a lyophilized fowl pox vaccine virus produced from a local isolate of Fowl Pox Virus (FPV) by National Veterinary Research Institute (NVRI) Vom, Nigeria. The hyper immune serum (positive test control) was obtained from two 10 weeks old pullets that were inoculated at Day 1 with 1mL of FP vaccine. Booster doses were administered at Day 8 and 12. Thereafter, serum was obtained at Day 21 post-vaccination tested for positivity against the vaccine antigen and preserved as positive control.

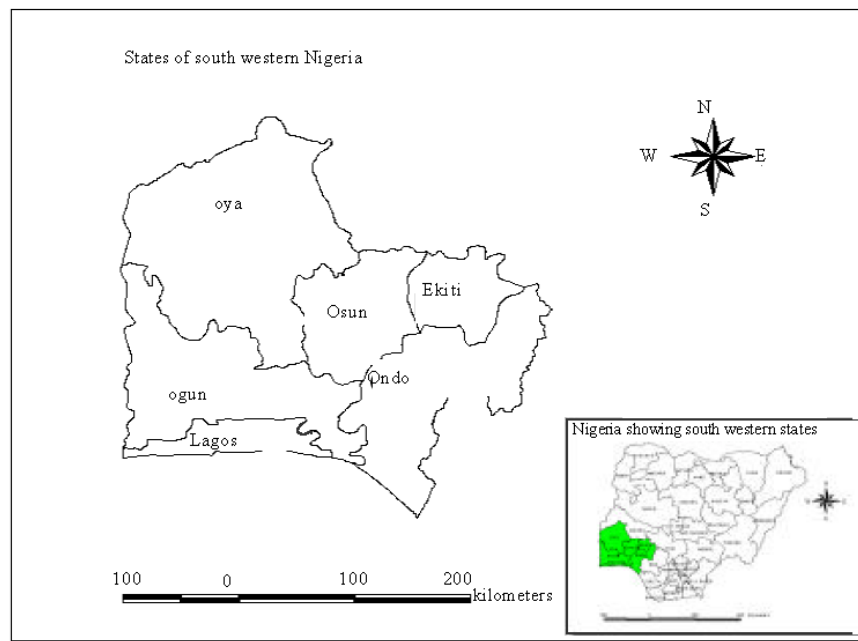


Fig. 1: Showing some of the states where samples were taken

The negative control serum was a negative chicken serum produced by IDDEX Laboratories ® USA. The least concentration (highest dilution) of reagents that gave the least optical density for the negative control and the greatest difference between the positive and negative controls were regarded as the optimal working dilutions.

**Elisa procedure:** The ELISA procedure was conducted essentially by adaptation of the method described earlier (Ohore *et al.*, 2002) for fowl typhoid with some modifications. The optimal dilutions obtained following the chequer board titration for indirect Fowl pox ELISA at Antigen1/5,000, Serum1/50, Conjugate 1/10,000.

**Test validation:** The test was considered valid only when the difference between the mean optical densities of the positive and negative control samples was greater than 0.30, in addition to the mean optical density value of the negative control being less than or equal to 0.35.

**Data presentation and analysis**

**Mean optical densities and samples to positive ratios:** The Samples to Positive Ratio (SPR) were determined from the mean optical densities of the test serum samples based on the standard formula previously described (Ohore *et al.*, 2002). The data were analysed using the paired t test of comparing means and determines differences between the type of bird.

**Prevalence:** To obtain the prevalence, the cut-off value for the samples to positive ratio was determined by the formula:

$$\text{Cut of value} = \frac{\text{NCX}}{\text{PCX} - \text{NCX}}$$

Where,

Ncx : Mean optical density of negative control

Pcx : Mean optical density of positive control

Samples with values of SPR grater than the cut-off were designated as positive.

Otherwise, it was designated as negative. The COV represents samples with SPR equal to twice the values for the negative control.

The prevalence per group of bird was calculated thus:

$$\text{Prevalence}(\%) = \frac{\text{Number of positivesamplein group} \times 100}{\text{Total number of samplesin - group}}$$

**RESULTS**

**Antibody titres:** The seroprofile of antibodies to FPV among different types of vaccinated and non-vaccinated chickens sampled are shown in Table 1 and 2. An overall

Table 1: Sero-prevalence of fpv antibodies among different types of non-vaccinated chickens in southwestern Nigeria

Type of bird	Farm location	No. of samples	No. positive (%)	Mean Sp ratio
Local				
Chicken	Oyo State	184	164 (89)	1.346±0.134
Growers	Oyo state	20	2 (10)	1.234
Layers	Oyo State	40	30(75)	1.115±0.113
Breeders	Lagos State	60	48 (80)	1.155±0.089
All birds	Total	304	244 (80)	

Table 2: Sero-prevalence of antibodies to FPV among different types of vaccinated birds

Type of bird	No. of Farm	samples	No. positive (%)	Mean OD +SD
Layers	Farm A Ogun state	90	42 (93)	1.154±0.088
	Farm B, Ondo state	64	31 (97)	1.245±0.084
Breeders	Abeokuta, Ogun State	94	94 (100)	1.187±0.105
All birds	Total	248	240 (97)	

prevalence of 80% was observed for all the non-vaccinated poultry samples obtained form four states of south-western Nigeria.

The local chicken had the highest antibody titres (1.351±0.134) among all the birds and also showed the widest range and standard deviation in titres. Among the exotic chickens, the highest titre of 1.245±0.088 was observed in vaccinated layers in Farm B while the lowest titres were seen in the non-vaccinated exotic birds.

Using the paired t-test, there was a significant difference in titres (p<0.001) between the local chicken group and the commercial chickens. The non-vaccinated breeder group was significantly different in titres (p<0.05) from every other group except the non-vaccinated layer group, while the non-vaccinated layers were only significantly different (p<0.001) from the local chickens. There was no significant difference between both types of vaccinated chickens (p<0.05).

**DISCUSSION**

The overall prevalence of fowl poxvirus antibodies in non-vaccinated poultry in this study was 80%. This is remarkably high, compared to the prevalence of 97% obtained in the vaccinated chickens indicating a high activity of fowl poxvirus in poultry in south-western Nigeria. Among the non-vaccinated types of birds, a prevalence of 89% was seen in local chickens, 10% in growers, 75% in layers and 80% in breeders. In an earlier survey following outbreaks of the disease, fowl pox was observed in 8 of 158 flocks (5%) of local chickens in Zaria (Saidu *et al.*, 1994).

There is little documentation on the epidemiology of fowl poxvirus infection in Nigeria and there has been no previous report of the use of the ELISA in the serodiagnosis of fowl pox in the indigenous chicken. In this study, the ELISA was observed to be a very useful

tool in detection of antibodies to field exposure to FPV in poultry and in the assessment of humoral response to vaccination.

The antibodies observed in non-vaccinated birds would either be due to active infection with the FP virus as there was no vaccination in these birds or due to recent recovery from the clinical disease. Because of the genetic makeup and inherent stability of the fowl poxvirus, it has been observed to persist in cabs and in the environment, which may be the source of infection (Tripathy and Reed, 2003). Although clinical disease was not observed in these birds, fowlpox has been shown to be an important disease of poultry in Nigeria (Saidu *et al.*, 1994).

The prevalence among the non-vaccinated birds was higher in the local chickens than the exotic chickens probably due to the higher virus activity in local birds, which may be due to the free-range management that exposes the birds to a wide range of pathogens in its environment. Similar observations have also been made with other pathogens in this group of birds e.g., Egg drop syndrome (Adedokun and Durojaiye, 1999) Newcastle disease (Ohore *et al.*, 2003) Fowl typhoid (Ohore *et al.*, 2002) and Mycoplasmosis (Abdu *et al.*, 1983).

The mean±S.D of the titres in the local birds (1.351±0.134) was significantly higher than all the other groups including the vaccinated birds ( $p<0.001$ ) indicating that these birds respond very well immunologically to the virus and the fact that the bird are well adapted to adverse tropical conditions unlike the exotic poultry (Mark *et al.*, 1969). However, it is not known if there is any genetic basis for the higher response although the local chicken has been shown to have a higher bursal index when compared to commercial chickens (Aire and Ojo, 1974; Okpe, 2001).

The wide variation in antibody titres in these birds is probably due to the varied conditions each bird is exposed to as a result of differences in the time and dose of exposure as well as phase of immune response as opposed to the near uniform exposure in intensive management system under which exotic birds that were vaccinated were raised. It may also be due to different rates of immune response in these birds, with some being high responders and other low responders as reported by Msoffe *et al.* (2001).

The growers had significantly lower prevalence of 10% compared to the other groups. Although FPV can occur in all ages of birds, younger birds are less susceptible to clinical diseases owing to their poorly developed combs and wattle. The low prevalence may be related to their age, as there is lesser chance of exposure to the virus as compared to the older birds that may have been continually exposed to the virus. This finding was in

contrast to those of Saidu *et al.* (1994) that reported clinical fowlpox disease in birds less than 10 weeks of age with about 60% of the outbreaks occurring in 2weeks old chicks with high mortalities. The absence of antibodies in these young birds may have conferred a state of low resistance and this may predisposed them to the clinical disease especially as they had contact with the older infected birds.

The non-vaccinated layers and breeders showed sero-prevalence of 75 and 80%, respectively. These are both high and further show the relative endemicity of fowl poxvirus infection in Nigerian commercial poultry. The antibodies observed in these birds may be due mechanical transmission from insects such as mosquitoes or exposure to dried infected scabs and aerosol containing fowl poxvirus particles. The high activity among the breeders is particularly significant because infection in these birds can result in decreased egg production and impaired fertility (Tripathy and Reed, 2003).

Among the vaccinated exotic poultry, sero-prevalence of 95 and 100% were observed in layers and breeders, respectively. There were cases of negative samples among the layers, 7% in farm A and 3% in farm B. This finding buttresses the need for sero-monitoring of vaccinated flocks. In recent years, outbreaks of fowl pox have been reported in the US in previously vaccinated chickens (Fatumbi and Reed, 1996; Singh *et al.*, 2000) while in Nigeria, there had been field reports of post vaccination outbreaks.

The mean titres in vaccinated layers (1.192) and breeders (1.187) were not significantly higher than the non-vaccinated layers (1.115) and breeders (1.155) ( $p<0.05$ ). However, it was significantly lower than for the local chicken (1351) ( $p<0.001$ ). The significantly high prevalence in the non-vaccinated poultry in Lagos and Oyo states with prevalence of 80 and 75%, respectively show the relative endemicity of the FPV infection in poultry in southwestern Nigeria.

In this study, the ELISA technique has been employed to provide data on the level of fowlpox virus activity in poultry in southwestern Nigeria. Although the distribution of fowl pox in commercial poultry is worldwide (Odend'hal, 1983) the incidence is variable and as shown in this study, may be affected by bio security measures and managerial practices on the farm (Tripathy and Reed, 2003).

The local chicken seems to be well adapted to exposure to FPV, as shown by the high titres of FPV antibodies in affected birds being significantly higher than even the vaccinated poultry. These well adapted chickens may be very important in the epidemiology of FPV by acting as potential reservoirs and carriers of

infection to the more susceptible exotic breeds in commercial enterprises as has been reported for other diseases (Adu *et al.*, 1986). This is particularly important in indigenous bird populations around commercial farms as mosquitoes play a very important role in the epidemiology of fowl pox. It is then imperative to stimulate the awareness of farmers on the need to vaccinate indigenous chickens. Further studies on the local chickens to determine genetic basis of resistance should also be encouraged. This could be useful in the production of disease resistance breeds for commercial poultry especially with recent advances in genomic research.

An effective national control against FP should take into consideration the role of the local chicken in the epidemiology of the disease. With emerging poultry diseases and more virulent strains of the known disease arising, diseases such as fowlpox could pose a threat to the poultry industry.

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