Research Note: Detection of infectious bursal disease virus antibodies in free-living wild birds in Zaria, Nigeria

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ABSTRACT Infectious bursal disease virus (**IBDV**) is an immunosuppressive pathogen of poultry causing great economic losses to the poultry industry. In this study, the IBDV antibodies were detected in captured free-living wild birds in Zaria, Nigeria. One hundred and fifty free-living wild birds, comprising 30 birds each of 5 different species, were sampled over a period of 9 months. Blood samples were collected from each bird, and harvested sera were tested for IBDV

antibodies using enzyme-linked immunosorbent assay. Results indicated IBDV seroprevalences in speckled pigeon (6.67%) and cattle egret (3.33%). In conclusion, the detection of IBDV antibodies in free-living wild birds in this study is indicative of previous natural exposure of these birds to the virus. These species of wild birds could therefore serve as carriers of these viruses and, consequently, transmit these viruses to chickens.

Key words: infectious bursal disease, wild bird, sera, seroprevalence, exposure

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INTRODUCTION

Infectious bursal disease (**IBD**) or Gumboro disease is an acute, highly contagious disease of chickens caused by infectious bursal disease virus (IBDV) (Eterradossi and Saif, 2008). The disease is characterized by bursal lesions, bursal atrophy, and immunosuppression in chickens between 3 wk and 3 mo of age (Mahgoub, 2012; Magwod et al. 2014). The virus responsible for IBD is a bisegmented, double-stranded RNA virus, belonging to the genus Avibirnavirus and family Birnaviridae (Baxendale, 2002; Jackwood et al. 2018). It shows selective tropism for lymphoid tissue and has affinity for immature B lymphocytes in the bursa of Fabricius (Lukert and Saif, 2003; Mwenda et al. 2018). It has been reported to also cause lymphoid depletion in the bursa of Fabricius in free-living wild birds, but infection is generally subclinical (AHA, 2009). The natural hosts of IBDV are domestic fowls including chickens and turkeys (Jackwood and Sommer-Wagner, 2007). The mode of transmission of IBDV is primarily through fecal-oral

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route, with aerosol spread considered to be less important (AHA, 2009). Two serotypes of IBDV (1 and 2) have been recognized with existence of considerable antigenic variation within each serotype (Motohiko et al. 1998; Jackwood et al. 2018). Serotype 1 is pathogenic only to chickens and can be further divided into classical virulent, attenuated, antigenic variant and very virulent IBDV based on their virulence and antigenicity (Wang et al. 2007; Mawgod et al. 2014; Jackwood et al. 2018). Serotype 2 viruses are naturally avirulent and do not cause clinical disease in chickens and turkeys (Motohiko et al. 1998). Serological evidence of serotype 1 IBDV infection in wild birds suggests that wild birds may play a role in the epidemiology of IBDV by serving as reservoirs for the virus (Gilchrist, 2005; AHA, 2009). Reports have shown that serotype 2 of IBDV is more prevalent in many species of free-living wild birds, with the natural host considered to be turkeys (Motohiko et al. 1998).

There is dearth of information on the IBDV status of free-living wild birds in Zaria, Nigeria. Therefore, the aim of this study was to detect IBDV antibodies in free-living wild birds in Zaria, Nigeria.

MATERIALS AND METHODS

Ethics Statement

The study protocol and all animal studies were approved by the Ahmadu Bello University Committee

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The authors declare that they have no conflict of interest.

Table 1. Seroprevalence of infectious bursal disease virus in free-living wild birds in Zaria,
 Nigeria, using indirect enzyme-linked immunosorbent assay.

Species of birds	Number of samples tested	Number of samples positive	Prevalence
Laughing doves	30	0	0.00
Speckled pigeons	30	2	6.67
Cattle egrets	30	1	3.33
Village weavers	30	0	0.00
African silverbills	30	0	0.00
Total	150	3	2.00

on Animal Care and Use (ABUCACU), Ahmadu Bello University, Zaria, Nigeria.

Sample Size

The sample size for the study was calculated using the following formula by Mahajan (1997):

$$N = Z^2 pq/d^2$$

where N = sample, Z = the appropriate value from the desired confidence (1.96), p = expected prevalence, q = 1 – Prevalence, d = allowable error.

Using relative seroprevalence of 6.15% from a previous study by Adamu et al. (2017),

 $N = 1.96^2 \times 0.0615 \times (1-0.0615)/0.05^2$

N = 88.69.

To increase the chances of detecting the antibodies, 150 blood samples and 150 sera were collected.

Sampling of Birds

Based on convenience sampling method, a total of 150 birds comprising 30 each of laughing doves (*Spilopelia* senegalensis), speckled pigeons (*Columba guinea*), cattle egrets (*Bubulcus ibis*), village weavers (*Ploceus cucullatus*), and African silverbill (*Euodice cantans*) were sampled alive over a period of 9 mo (March–December, 2017). These species of birds were used based on their availability at the particular time of capture. The birds were captured alive and unhurt using wooden traps kept at strategic positions around poultry houses located at different locations within the environ. The 10 poultry farms were selected based on frequency of visit of these free-living wild birds and previous history of IBD outbreaks.

The captured birds were identified by an ornithologist in the Department of Zoology, Faculty of Life Sciences, Ahmadu Bello University Zaria. Thereafter, physical examination was conducted on each bird to be sure there were no lesions, wounds, or ectoparasites.

Blood Sample Collection

Blood sample (0.5-1 ml) was collected from each bird via the wing vein using sterile hypodermic syringes and 23G needles. Sera were harvested from the blood, transferred into labeled sterile plastic containers, and stored at -20° C until used for IBDV antibody detection. After sample collection, each bird was marked to avoid repeated sampling and released into the environment.

Enzyme-Linked Immunosorbent Assay for Detection of IBDV Antibody

The 150 test sera were subjected to indirect ELISA following the manufacturers' instructions (IDEXX IBD-XR Ab Tests Technical Guide). IBD virus antibody test kit (designed for serotype 1 strain) used was obtained from IDEXX IBD-IDEXX Laboratories Inc., Westbrook, ME. The absorbance values were measured and recorded at 650 nm wavelength using an ELISA microtiter plate reader. The relative level of antibody to IBDV in the sample was determined by calculating the sample-to-positive (S/P) ratio. Sera with S/P ratios of less than or equal to 0.20 were interpreted as negative. Sample-to-positive ratios greater than 0.20 were interpreted as positive and indicated vaccination or exposure to IBD virus according to the manufacturers' technical guide.

Data Analyses

The ELISA data were presented as percentages in Table. The prevalence of antibodies to IBDV was calculated for each bird species using the formula outlined by Bennette et al. (1991).

RESULT

The result of this study shows seroprevalence of IBDV antibodies in speckled pigeons (6.67%) and cattle egrets (3.33%) but no serological evidence in laughing doves, village weavers, and the African silverbills in Zaria, North West Nigeria (Table 1).

DISCUSSION

In this region, vaccination of chickens against IBD is frequently carried out. Fagbohun et al. (2000) reported seroprevalence of IBDV antibodies in cattle egrets (20.0%) using ELISA at Ibadan, South West Nigeria. Also, antibodies to IBDV have also been reported in speckled pigeons (6.0%) and laughing doves (13.04%) in Kano using ELISA (Adamu et al. 2017).

From this study, it is believed that these species of free-living wild birds have been previously exposed to IBDV and could play important roles in the natural maintenance and spread of the virus in poultry because of their migratory potentials. The common IBD vaccines used in Nigeria and the surrounding areas are live vaccines from serotype 1 strain, and the wild birds that tested positive did not show signs of having been exposed to these vaccine strains.

The presence of IBDV antibodies in free-living wild birds in this study might have resulted from indirect interaction through frequent visits to commercial poultry farms and feeding around poultry houses in this region (Oladele et al. 2011). Also, these free-living wild birds have been found among the chickens in areas where local chickens are reared on free-range management system and around live bird markets. These interactions allowed for possible ingestion of the viruses by these birds, thus suggestive of the seroprevalence observed in this study. The detection of IBDV antibodies in free-living wild birds is indicative of previous natural exposure of these birds to the viruses at some point in their life. These free-living wild birds therefore may serve as carriers of these viruses after migration to poultry houses and possible dissemination of the virus to chickens.

It was observed that IBDV antibodies exist in free-living wild bird populations. The detection of IBDV (2.0%) antibodies in free-living wild birds in this study indicated previous natural exposure to these viruses. To the best of our knowledge, this is the first report on the serological studies of IBDV in these species of wild birds in Zaria, Nigeria.

Whether these viruses cause clinical diseases and pathology in free-living wild birds require further investigation. It is recommended that further research involving experimental inoculation and more species of birds should be carried out in Zaria, Nigeria, to understand the pathology, pathogenesis, epidemiology, and status of IBDV in wild bird populations.

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