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Short Communication

Seroprevalence of chicken infectious anemia virus infection among some poultry species in Maiduguri, Nigeria

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ABSTRACT

Objective: This study was designed to investigate the seroprevalence of Chicken Infectious Anemia Virus (CIAV) among selected poultry species in Maiduguri, Nigeria.

Materials and method: ELISA kit (X-Ovo Flockscreen™, Cat. No.V085 5 plates. February, 2014 - Xnew kit format), Chicken serum, enzyme conjugate reagent, adhesive cover, wash buffer, substrate reagent, stop solution. Serum samples from village chickens, broilers, layers, ducks, turkeys and geese in Maiduguri were tested for CIAV antibodies using Enzyme Linked Immunosorbent Assay (ELISA) as per the manufacturer's protocols at the Viral Research Laboratory, Faculty of Veterinary Medicine, University of Maiduguri, Nigeria. The results were presented in simple percentages, bar charts and analyzed using SPSS Version 16 software.

Results: Out of 944 sera from different species of poultry tested, an overall seroprevalence of 38.5% (n=363/944) was recorded in this study. The species distribution showed village chickens had 41.4% (n=166/944) prevalence, layers with 23.0% (n=12/52), broilers 46.6% (n=146/313), turkeys 23.6% (n=30/127), ducks 13.7% (n=4/29) and geese 22.7% (n=5/22) prevalence for CIAV antibodies.

Conclusion: The result of this study shows that CIAV infection is present among different poultry species in the study area and therefore highlight the need for continuous surveillance so as to control further spread of the virus.

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INTRODUCTION

Chicken infectious anemia (CIA) otherwise known as Anemia-dermatitis syndrome or blue wing disease (Rozypal et al., 1997) is an emerging viral infection of poultry (mostly 2-4 weeks old) characterised by anaemia, subcutaneous haemorrhage, immunosuppresion, cachexia and high mortality (Rozypal et al., 1997). It is caused by CIA virus (CIAV), belonging to the family *Circoviridae* having a single stranded DNA virus with icosahedral symmetry (Fenner et al., 1993). In 1974, the virus was first isolated from commercial chicken in Japan (Yuasa et al., 1979). The disease may cause economic loss particularly to the broiler industry and the specific pathogen free eggs producers (Schat, 2003; Oluwayelu, 2010). These losses are mainly due to poor growth, high mortality and cost of antibiotics used to control secondary bacterial infections (McNulty, 1991). Serological survey has indicated that CIAV infection is common throughout the world (Jordan and Pattison, 1998) and has been isolated in countries like Japan (Yuasa et al., 1979) Germany (Todd et al., 1992), Nigeria (Oluwayelu and Todd, 2008), Iran (Mahzaunieh et al., 2005), Argentina (Buscaglia et al., 1994), Israel (Davidson et al., 2004) and USA (Hoerr et al., 2005). In Africa, CIAV was first isolated from broiler chickens in South Africa (Witch and Maharaj, 1993) followed by Egypt (Aly, 2001; Hussein et al., 2002) and Nigeria (Oluwayelu et al., 2005). Despite the aforementioned economic threat of CIA to poultry industry and wide spread nature of the virus, there is dearth of information on the disease in the study area. The objective of this study therefore is to provide the seroprevalence information of CIAV among poultry species in the study area.

MATERIALS AND METHODS

Ethical statement: The study was conducted by following the international standards in terms of animal welfare and ethics.

Study area: This study was carried out in Maiduguri, Borno State, Nigeria. Maiduguri is the capital city of Borno State and lies between latitude 10.20°N and 13.40°N to the north, longitude 9.80°E and 14.40°E to the east and occupies an area of 69.436 square kilometers. Borno State shares international borders with Niger to the north, Chad to the north east and Cameroon to the east (Musa and Pindar, 2005).

Study population: Blood samples were collected from village chickens, broilers, layers, turkeys, ducks and geese.

Sampling and storage: A non-probability convenient sampling was used in this study. A total of nine hundred and forty four (944) blood samples were collected and directly dispensed into labeled plain vacutainer tubes and allowed to clot. After clotting, the samples were spun at 1,500 rpm using a centrifuge and the sera were harvested, stored in an appropriately labeled cryotubes and kept at -20°C until tested.

Serology: Sera from the different poultry species were tested for the presence of CIAV antibodies using ELISA kit (X-Ovo Flockscreen™ Cat. No. V085 5 plates. February, 2014 - Xnew kit format). The ELISA test was carried out according to the manufacturer's protocols in Viral Research Laboratory, Faculty of Veterinary Medicine, University of Maiduguri, Nigeria. Briefly, 50 µL of a 1:500 dilution of each test serum was added to a well of microtiter plates precoated with CIAV antigen. Each test sample was run in a single well and the positive and negative controls were run in duplicates. Each plate was covered with an adhesive cover and incubated at 37°C for 60 min. The adhesive cover on each plate was removed and the plates were washed four times with the supplied wash buffer and the plates were inverted and tapped firmly on absorbent paper.

Fifty microlitres (50 µL) of enzyme conjugate reagent (alkaline phosphatase labeled rabbit anti-chicken IgG in tris butter with an inert blue dye and sodium azide 0.1% v/v) was added to each well, mixed by gently tapping the side of the plate. The plates were covered with the adhesive cover and incubated at 37°C for 60 min. Again the adhesive covers were removed and plates were washed 4 times with wash buffer. Each washed plate was inverted and tapped firmly on absorbent paper. Finally, 50 µL of substrate reagent (phenolphthalein mono-phosphate and enzyme cofactors in a diethanolamine buffer) was added to each well of the plate and mixed by tapping the side of the plates. The plates were covered with the adhesive cover and incubated at 37°C 30 min. The adhesive cover was removed and 50µ1 of the stop solution (sodium hydroxide and a chelating agent in a diethanolamine buffer) was added to each well and mixed by gently tapping the side of the plate to obtain full color development and the plates were immediately read using microtiter plate reader at 550 nm.

Data analysis: Data obtained from the study were presented in simple percentages, bar charts and analyzed using SPSS Version 16 software. *P*-value less than or equal to 0.05 was considered statistically significant.

RESULTS

The ELISA result of the test sera of different species of birds in Maiduguri for CIAV antibodies (IgG) revealed an overall seroprevalence of 38.5%**.** The species distribution of the CIAV positive samples showed village chickens

166/401 (41.4%), layers 12/52(23.0%), broilers 146/313 (46.6%), turkeys 30/127(23.6%), ducks 4/29(13.7%) and geese 5/22 (22.7%) were positive for CIAV antibodies (**Table 1**). The sex distribution of the CIAV seropositive samples showed and overall prevalence of 41% among males and 32.9% among females (**Table 2**). The distribution of the OD values of the samples positive for CIAV antibody indicated the village chickens have 60.8% in lower OD values (0.3 - <10), 22.3% with middle OD values (10-<25) and 16.9% with high OD values (25-<40) (**Figure 1**).This was followed by the layers with 91.7% lower OD values and 8.3% middle OD values; and all the remaining birds reacted with 100% in lower OD values.

Table 1: Distribution of CIAV ELISA antibodies among different poultry species in Maiduguri, Nigeria

Poultry Type	Total tested (N)	Postive (%)
Village chicken	401	166 (41.4)
Layers	52	12 (23)
Broilers	313	146 (46.6)
Turkeys	127	30 (23.6)
Ducks	29	4 (13.7)
Geese	22	5 (22.7)
Total	**944**	**363 (38.5)**

P<0.05

Table 2: Sex distribution of seroprevalence of CIAV infection among different species of poultry in Maiduguri, Nigeria

Poultry Type	Males		Female	
	Tested (N)	+ (%)	Tested (N)	+ (%)
Village chicken	251	96(38.2)	150	70 (46.7)
Layers	NA*	NA*	52	12 (23.1)
Broilers	313	146(46.6)	0	0(0)
Turkeys	57	18(31.6)	70	12 (17.1)
Ducks	15	3(20.0)	14	1(7.1)
Geese	7	1(14.3)	15	4(26.6)
Total	**643**	**264(41.0)**	**301**	**99(32.9)**

P<0.05; NA= Not Applicable

DISCUSSION

Despite the economic importance of CIAV to the poultry industry, there is scanty of information on its prevalence in the study area. The overall CIAV seroprevalence rate of 38.5% observed might be attributed to natural infection since there was no vaccination against the disease in Nigeria. The overall CIAV seroprevalence of 38.5% in this study was similar to the 36.7% in Central African Republic and 34.9% in Cameroon, as reported by Snoeck et al. (2012) The overall seroprevalence rate (38.5%) observed in this study was lower than the 55%, as reported in Western Nigeria by Owoade et al. (2004); 88.9% among apparent healthy chickens in southwestern Nigeria (Owoade et al., 2004); 66.2% in backyard chickens (Oluwayelu and Todd, 2008); 86.1% among apparently healthy chickens (Oluwayelu et al., 2009) all in western Nigeria; 59% among commercial chickens in Zaria northern Nigeria (Okpanachi, 2015); 87.7% among commercial chickens in Iran (Mahzaunieh et al., 2005). The difference between the present study and those reported by other researchers could be due to number of other species of poultry included in the present study. To support this, Snoeck et al., (2012) reported that seroconversion is generally not homogenous even within a flock nor between species. This, therefore tends to complicate comparison between studies.

Broilers recorded the highest seroprevalence rate of 46.6% when compared to other poultry species studied. This disagrees with Okpanachi (2015) who reported higher prevalence of the disease in laying chicken compared with broilers.

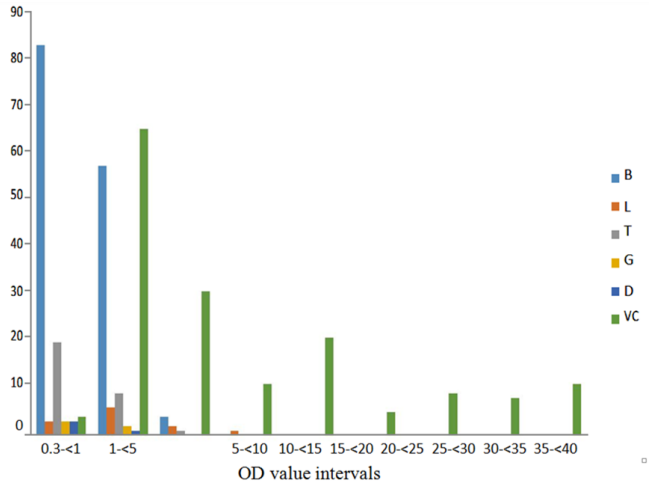

Figure 1: Bar chart showing the optical density (OD) values of CIAV ELISA antibody positive Serum samples. B=broilers, L=layers, T=turkeys, G=geese, D=ducks, VC= village chickens. Optical density value less than 0.306 is considered low OD value, while OD values equals to 0.306 are considered middle, and OD values greater than 0.306 are considered high OD values.

Apart from reports on prevalence of CIAV in village chickens and commercial chickens (layers and broilers) in Nigeria, no documented report on the prevalence of the virus among other poultry species (ducks, turkeys and geese) were made in the study area. The present study revealed CIAV prevalence among turkeys (23.6%), ducks (13.7%) and geese (22.7%), which are raised together alongside the chickens. This suggests that this group of birds could probably serve as potential reservoirs of this virus as there is no evidence of clinical CIA in these species of birds reported. A higher prevalence rate of 41.0% of the virus was recorded in male than in female (32.9) poultry from this study. This is similar to what was reported by Lawal et al. (2014), who reported higher prevalence in male (59.9%) than female (52.2%) birds.

The higher prevalence in males in the present study could be due to the fact that male birds were sampled more than the female birds because more of the male birds are sold out and slaughtered at live bird markets and houses while the female birds are kept for breeding purposes.

CONCLUSION

It was observed in this study that CIAV infection was 38.5% among the different poultry species studied in Maiduguri, Nigeria. It's recommended that further research be carried out on the pathology, pathogenesis, epidemiology and molecular studies of CIAV among different poultry species in the study area so as to understand the detailed molecular epidemiology of the virus in other to prevent it.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

AUTHORS' CONTRIBUTION

YMS, ADE and MBA carried out ELISA procedure. TMH drafted the manuscript. SSB and DOO polished English of the manuscript. MYZ and MMM participated in the study design and its coordination. All authors read and approved the final manuscript.

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