Survey for Newcastle Disease Virus Antibodies in Local Chickens, Ducks and Pigeons in Makurdi, Nigeria

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Abstract: This study aimed to determine the seroprevalence of Newcastle disease (ND) among some local poultry species from live bird markets (LBM) and households in Makurdi Benue State, Nigeria. A total of 543 (300 local chickens, 199 pigeons and 44 ducks) sera samples were collected between October 2018 to April 2019. Sera samples were tested using Haemagglutination inhibition (HI) test. The overall seroprevalence of Newcastle disease virus (NDV) antibodies was 9.9% (54/543). The prevalence of antibodies to ND was found to be higher in ducks 20.5% (9/44), followed by local chickens 10% (30/300) and pigeons 7.5% (15/199) respectively. There was statistical significant association (P = 0.000817) between location of sampling of local poultry and seropositivity for ND. Also there was a statistical significant association (P = 0.0348) between species of local poultry and ND antibody titre. This study revealed that local chickens, ducks and pigeons in Makurdi could play significant role in the epidemiology and transmission of ND to susceptible commercial exotic birds or other local poultry species especially when reared in close proximity. The study concluded that the prevalence of ND antibodies indicated the presence of the virus among the population and ducks and pigeons could be reservoirs and carriers for NDV. Vaccination campaign should be organized for local poultry species for the control of ND in the study area.

Keywords: Newcastle Disease Virus, Antibody Titre, Local Poultry, Haemagglutination Inhibition Test, Live Bird Market

1. Introduction

Newcastle disease (ND) is a major viral disease of economic importance in poultry and rated as one of the greatest constraints to the development of rural poultry production in Nigeria and in most developing countries [1]. The disease is caused by virulent strains of avian paramyxovirus type 1 (AMPV-1) serotype of the genus *Avulavirus* belonging to the family *Paramyxoviridae*. The paramyxoviruses isolated from avian species have been classified by serological testing and phylogenetic analysis into ten subtypes designated APMV-1 to APMV-10 [2]. According to the severity level of the outbreak in chicken, Newcastle disease virus (NDV) was classified into three pathotypes namely: lentogenic, mesogenic, and velogenic strains. Velogenic strain is distinguished into neurotropic and viscerotropic form [3]. The disease is characterized by gastrointestinal and respiratory signs often associated with nervous disorders and high mortality (up to 100%). Several species of birds are affected by ND with chickens and turkeys being readily susceptible. Over 250 species of birds have been reported to be susceptible to NDV as a result of natural or experimental infections, and it is likely that many more susceptible species exist but have not yet been identified [4]. In domestic pigeons, NDV is of high economic importance and worldwide occurrence has been reported including India [5, 6]. The ability of non-poultry avian species to introduce and disseminate NDV has been previously established, and feral pigeons (Columba spp.) have been implicated as carriers of NDV [7, 8]. The first report of ND in Nigeria was in 1952 [9], thereafter several cases have been reported in commercial, rural scavenging, captive and free-living wild birds making it enzootic across the entire country [10-13]. Serological evidence of ND infection in pigeons in Nigeria has been reported [14]. The poultry industry in Nigeria offer the quickest supply of animal protein to man in form of high quality eggs and meat and provides comparatively faster return to investment than...
cattle, small ruminants and pigs [15]. As practiced in most developing economies, poultry production systems are generally categorized into two major groups, namely; subsistent (free-range) and commercial poultry [16]. In Nigeria free range poultry accounts for over 80% of the poultry population [17]. The sustainability of this subsector is being threatened as a result of incessant outbreaks of ND in unvaccinated flocks and sporadically in vaccinated flocks [18].

Assessment of economic impact due to ND is not only limited to high mortality recorded from the outbreaks and the cost of control measures but also on the trade restrictions placed on the localities where outbreaks have been reported [19, 20]. Economic and financial losses as a result of incessant ND outbreaks in Nigeria are not being regularly quantified. An estimated 78,526 outbreaks of the disease were reported in 2008 across Nigeria with an estimated financial burden of 8.9 billion naira for local chickens alone [21]. The free-range chickens have been implicated in harbouring velogenic strains of the virus which have been considered a threat to the commercial poultry [10]. Available information on ND and its extent of distribution in local poultry species especially pigeons and ducks in Makurdi are scanty. This present study therefore aimed at detecting antibodies to Newcastle disease virus infection from some local poultry species sampled from households and major live bird markets in Makurdi, Benue State.

2. Materials and Methods

2.1. Study Area

The study was carried out in Makurdi the capital of Benue State, Nigeria. Makurdi is located in the North Eastern part of Benue State and lies on latitude 7°30'N and longitude 8°35'E. It is located within the flood plain of lower River Benue valley [22]. The physiographic characteristics span between 73-167m above sea level. The town is divided by the River Benue into the North and South banks, connected by two bridges: the railway bridge and the dual carriage bridge [22]. The study locations were four live bird markets (LBMs) and households within Makurdi metropolis. The locations were; North bank market, Wadata market, Wurukum market, Modern market and birds also sampled from households in the same locations.

2.2. Sampling Method and Sample Size

Simple random and convenient sampling methods were used for survey locations. The sample size was calculated using the formula of Thrustfield [23] using 1.6% anticipated prevalence obtained for ND virus in ducks in Lafia, Nasarawa State by [24], in pigeons 48.7% by [25] in Zaria, Kaduna State and in local chickens 45.6% by [26] in Benue State. A total of 543 sera were collected.

2.3. Collection of Blood

Blood sample was collected from each bird after proper restraint with the use of a 21 gauge needle attached to a 5 ml syringe to withdraw 1-2 ml of blood from the brachial vein. The blood collected was allowed to stand for two hours at room temperatures for clotting to occur and the serum decanted into a bijou bottle. The samples were then labeled and stored under ice then transported to the laboratory and kept at -20°C until used.

2.4. Detection of Newcastle Disease Antibodies

The LaSota strain of NDV and antibody positive serum were obtained from the National Veterinary Research Institute (NVRI), Vom and used as positive antigen and control serum respectively. A 1% suspension of chicken red blood cells (RBC) was prepared and used as indicator in the haemagglutination (HA) and haemagglutination inhibition (HI) tests according to methods described in OIE protocol [27]. Sera with HI antibody titre ≥1 log2 and ≥4 log2 were considered positive and protective respectively based on OIE manual [27].

2.5. Data Analysis

The data obtained from serology was subjected to SPSS package version 16. Categorical variables were evaluated using Chi-square to check for independence. Values of $P < 0.05$ were considered significant.

3. Results

An overall seroprevalence of 9.9% (54/543) was recorded among the different species of poultry examined. Out of the 300 local chicken sera sample tested 30 (10%) were positive for ND antibodies, while out of 44 ducks and 199 pigeon sera tested 9 (20.5%) and 15 (7.5%) were positive for ND antibodies respectively. There was a statistical significant association $P \leq 0.05$ ($P = 0.0348$; $X^2 = 6.715$) between species of local poultry and ND antibody titre (Table 1).

The seroprevalence and distribution of ND antibody titre by Haemagglutination inhibition test showed that 80% of the ducks sampled had antibody titre between 1-3 log2 while 0% and 20.5% had antibody titre ≥4log2 and ≥7log2 respectively. In local chicken, 92% had antibody titre between 1-3log2, 4% had antibody titre ≥4log2, and ≥7log2. A total of 91% had antibody titre between 1-3log2, 2.4% had antibody titre of ≥4log2, and 6.4% had antibody titre ≥7log2 (Table 2).

Table 1. Prevalence of Newcastle Disease Virus Among Poultry Species in Makurdi, Benue state, Nigeria.

<table>
<thead>
<tr>
<th>Poultry Species</th>
<th>No. of Sera Tested</th>
<th>(%) Sera Positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ducks</td>
<td>44</td>
<td>9 (20.5)</td>
</tr>
<tr>
<td>Pigeons</td>
<td>199</td>
<td>15 (7.5)</td>
</tr>
<tr>
<td>Local chickens</td>
<td>300</td>
<td>30 (10.0)</td>
</tr>
<tr>
<td>Total</td>
<td>543</td>
<td>54 (9.9)</td>
</tr>
</tbody>
</table>

$P = 0.0348$, $X^2 = 6.715$
The distribution of antibody titre according to sampling locations revealed that live bird market had a higher seroprevalence 11.1% (23/207) than birds sampled from different households 9.2% (31/336). Birds from LBM in North bank had seroprevalence of 3.1% (1/32), Wadata LBM had 35% (7/20), Modern Market had 14.3% (10/70) and Wurukum LBM had 5.9% (5/85). There was a statistically significant association (p-value = 0.000817, X^2 =18.914) between location of sampling of local poultry and seroprevalence for ND (Table 3).

The overall seroprevalence of Newcastle disease in the study area was 9.9%. None of the birds sampled had a history of previous vaccination against ND. Antibodies detected may be a result of natural infection since vaccination of the local poultry is rarely undertaken in Nigeria [28]. The prevalence rate was higher in ducks (20.5%) compared to chickens (10%) and pigeons (7.5%). Ducks are known carriers of the avirulent NDVs which may mutate to virulent ND virus strains [29]. Waterfowls such as ducks and geese may be infected but show few or no clinical signs even to the strains that are virulent to chickens [30]. The risk of NDVs transmission from ducks or water birds may be either through direct or indirect contact with poultry flocks such as contaminated feed and water or infection of wild bird species at common water sites and then transmission of the infection into poultry flocks [31]. This suggest that ducks in the study area should be considered as an important factor in the epidemiology of the disease. The seroprevalence recorded in local chickens in this study was 10%. This result is lower than the prevalence of 46% reported in village chickens in Borno State [32] and 54.67% in Nasarawa State [24] among local scavenging chickens kept under traditional management system. This variation could be due to differences in study settings or by exposure to mild virus strains that induced immunity but did not kill many chickens. Also, seroprevalence rate of ND antibodies could be attributed to factors such as the management system in traditional production where birds of different ages and species scavenge and interact together which may serve as a stress factor and favour infection [33]. The seroprevalence of 7.5% recorded in pigeons in this study is lower than 48.7% reported in Gombe State, Nigeria [34]. Although there was no report of clinical outbreak of ND in domestic pigeons in Nigeria, there has only been serological evidence of ND infections in pigeons in Nigeria [14]. Pigeons can be infected with ND virus and may therefore serve as reservoir of ND virus for more susceptible poultry species [25]. Feedstuffs contaminated with faeces of feral pigeons infected by pigeon paramyxovirus (PPMV) were considered to be sources of infection in many outbreaks, since pigeons easily come in contact with free-range birds. Thus, they may disseminate NDV among many different avian species [35]. There was an outbreak of ND in UK due to contamination of poultry feed with faeces of pigeons [36]. This implies that ND infected pigeons can play a significant role in transboundary transmission of the disease since they can fly several miles across regions. The distribution of the percentage of birds with HI protective antibody titre level (4Log2) was low compared with birds with detectable antibody level. This finding of NDV antibodies in these apparently healthy birds suggests that the birds have either recovered from clinical ND or are having subclinical infections [37]. This also implies that a greater proportion of the local poultry specie in the study area (91%) will be more vulnerable to challenge with a virulent strain of NDV if there is an outbreak of the disease because of low level of antibody against ND. Different antibody titre cutoff values has been used by different researchers for the interpretation of HI test results. This could be another reason for variation between studies, some authors considered an HI antibody titer ≥ 1log2 as positive [38, 39], whereas others used cut-off titers of ≥3log2 [40, 41]. However, the present study used ≥4log2 as HI protective antibody level (OIE) [42], which is similar with the cutoff values used by Gutierrez-Ruiz [43]. The present study revealed that the origin of the birds was significantly associated with ND seropositivity (p=0.05). Birds from Live bird markets had a higher prevalence rate when compared with those sampled from households. This may be due to the fact that live bird markets contribute to the persistence and spread of ND virus [44, 26]. These birds are exposed to birds from multiple sources having a higher tendency of circulating the virus and may serve as a source of infection to house hold chickens when introduced [45]. Other studies conducted on village chickens at live bird markets in

### Table 2. Seroprevalence and distribution of Newcastle Disease antibody titre by haemagglutination inhibition (HI) test in poultry species in Makurdi, Benue state, Nigeria.

<table>
<thead>
<tr>
<th>Species</th>
<th>% of birds with HI titre ≤3Log2</th>
<th>% of birds with HI titre ≥4Log2</th>
<th>% of birds with HI titre ≥7Log2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Duck</td>
<td>80 (35/44)</td>
<td>0 (0/44)</td>
<td>20.5 (9/44)</td>
</tr>
<tr>
<td>Pigeon</td>
<td>93 (184/199)</td>
<td>0.5 (1/199)</td>
<td>7 (14/199)</td>
</tr>
<tr>
<td>Local chicken</td>
<td>92 (276/300)</td>
<td>4 (12/300)</td>
<td>4 (12/300)</td>
</tr>
<tr>
<td>Total</td>
<td>91 (495/543)</td>
<td>2.4 (13/543)</td>
<td>6.4 (35/543)</td>
</tr>
</tbody>
</table>

### Table 3. Distribution of Newcastle disease antibody titre according to sampling locations in Makurdi, Benue state, Nigeria.

<table>
<thead>
<tr>
<th>Location</th>
<th>No of samples</th>
<th>No of positive</th>
<th>Prevalence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Northbank LBM</td>
<td>32</td>
<td>1</td>
<td>3.1</td>
</tr>
<tr>
<td>Wadata LBM</td>
<td>20</td>
<td>7</td>
<td>35</td>
</tr>
<tr>
<td>Modern market</td>
<td>70</td>
<td>10</td>
<td>14.3</td>
</tr>
<tr>
<td>Wurukum LBM</td>
<td>85</td>
<td>5</td>
<td>5.9</td>
</tr>
<tr>
<td>House hold</td>
<td>336</td>
<td>31</td>
<td>9.2</td>
</tr>
<tr>
<td>Total</td>
<td>543</td>
<td>54</td>
<td>9.9</td>
</tr>
</tbody>
</table>

P = 0.000817, X^2=18.914

### 4. Discussion

The overall seroprevalence of Newcastle disease in the study area was 9.9%. None of the birds sampled had a history of previous vaccination against ND. Antibodies detected may be a result of natural infection since vaccination of the local poultry is rarely undertaken in Nigeria [28]. The prevalence rate was higher in ducks (20.5%) compared to chickens (10%) and pigeons (7.5%). Ducks are known carriers of the avirulent NDVs which may mutate to virulent ND virus strains [29]. Waterfowls such as ducks and geese may be infected but show few or no clinical signs even to the strains that are virulent to chickens [30]. The risk of NDVs transmission from ducks or water birds may be either through direct or indirect contact with poultry flocks such as contaminated feed and water or infection of wild bird species at common water sites and then transmission of the infection into poultry flocks [31]. This suggest that ducks in the study area should be considered as an important factor in the epidemiology of the disease. The seroprevalence recorded in local chickens in this study was 10%. This result is lower than the prevalence of 46% reported in village chickens in Borno State [32] and 54.67% in Nasarawa State [24] among local scavenging chickens kept under traditional management system. This variation could be due to differences in study settings or by exposure to mild virus strains that induced immunity but did not kill many chickens. Also, seroprevalence rate of ND antibodies could be attributed to factors such as the management system in traditional production where birds of different ages and species scavenge and interact together which may serve as a stress factor and favour infection [33]. The seroprevalence of 7.5% recorded in pigeons in this study is lower than 48.7% reported in Gombe State, Nigeria [34]. Although there was no report of clinical outbreak of ND in domestic pigeons in Nigeria, there has only been serological evidence of ND infections in pigeons in Nigeria [14]. Pigeons can be infected with ND virus and may therefore serve as reservoir of ND virus for more susceptible poultry species [25]. Feedstuffs contaminated with faeces of feral pigeons infected by pigeon paramyxovirus (PPMV) were considered to be sources of infection in many outbreaks, since pigeons easily come in contact with free-range birds. Thus, they may disseminate NDV among many different avian species [35]. There was an outbreak of ND in UK due to contamination of poultry feed with faeces of pigeons [36]. This implies that ND infected pigeons can play a significant role in transboundary transmission of the disease since they can fly several miles across regions. The distribution of the percentage of birds with HI protective antibody titre level (4Log2) was low compared with birds with detectable antibody level. This finding of NDV antibodies in these apparently healthy birds suggests that the birds have either recovered from clinical ND or are having subclinical infections [37]. This also implies that a greater proportion of the local poultry specie in the study area (91%) will be more vulnerable to challenge with a virulent strain of NDV if there is an outbreak of the disease because of low level of antibody against ND. Different antibody titre cutoff values has been used by different researchers for the interpretation of HI test results. This could be another reason for variation between studies, some authors considered an HI antibody titer ≥ 1log2 as positive [38, 39], whereas others used cut-off titers of ≥3log2 [40, 41]. However, the present study used ≥4log2 as HI protective antibody level (OIE) [42], which is similar with the cutoff values used by Gutierrez-Ruiz [43]. The present study revealed that the origin of the birds was significantly associated with ND seropositivity (p=0.05). Birds from Live bird markets had a higher prevalence rate when compared with those sampled from households. This may be due to the fact that live bird markets contribute to the persistence and spread of ND virus [44, 26]. These birds are exposed to birds from multiple sources having a higher tendency of circulating the virus and may serve as a source of infection to house hold chickens when introduced [45]. Other studies conducted on village chickens at live bird markets in...
Nigeria by [46, 44, 47]) showed 35.8%, 25.5% and 65.1% seroprevalent rates respectively. Furthermore, contact of birds of one rural area with those of another rural area through gift and sale of rural chickens in which in some cases are diseased or carriers of some diseases may facilitate the spread NDV among flocks [48].

5. Conclusion

The findings of this research work revealed the presence of antibodies against ND Virus in sera of apparently healthy local chickens, ducks and pigeons in Makurdi, Benue State Nigeria. The overall seroprevalence in ducks (20.5%) was higher than that of the local chickens (10%) and pigeons (7.5%) respectively. This finding is not in agreement with the general belief that local chickens usually have a higher value of NDV antibody than other domestic poultry species. Local poultry could play a significant role in the epidemiology and transmission of the NDV to exotic and commercial poultry species especially when reared in close proximity. It is therefore recommended that routine vaccination programmes against ND should include other poultry species such as ducks and pigeons to prevent disease outbreak that could lead to economic loss in the poultry industry.

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References


