ISSN 1682-8356
ansinet.org/ijps

INTERNATIONAL JOURNAL OF

# POULTRY SCIENCE

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International Journal of Poultry Science 13 (5): 299-303, 2014
ISSN 1682-8356


# Seroprevalence of Five Important Viral Diseases in Commercial Chickens in Grenada, West Indies

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**Abstract:** This study was conducted to estimate the seroprevalence of New Castle disease (ND), Infectious bursal disease (IBD), Chicken infectious anemia (CIA), Avian Pneumovirus (APV) and Avian influenza (AI) in commercial chickens in Grenada, West Indies. In total, 226 and 233 sera were collected, respectively from layer (6 to 18 months age) and broiler (6 to 7 weeks of age) chickens in six regions (parishes) of Grenada and were tested using commercial ELIZA kits. The overall seroprevalence of ND, IBD, CIA, APV and AI was 47.7% (95% CI; 41.2-54.2%), 34.7% (95% CI, 28.4-40.9%), 92.9% (95% CI; 89.5-96.2%), 61.9 (95% CI, 55.6-68.2%) and 0%, respectively for layers and 48.25 (95% CI, 41.85-54.6%), 40.35 (95% CI,34.0-46.6%), 58.3% (95% CI, 52.0-64.2%), 31.7% (95% CI.25.7-37.7%) and 0%, respectively for broilers. The presence of antibodies for more than one disease agent in the same chicken indicates that several pathogens are circulating within the same premises. This survey shows that major poultry diseases are threatening the productivity of commercial chickens in Grenada. This is the first report on disease surveillance in commercial chickens in Grenada.

**Key words:** Antibodies, commercial chickens, Grenada, viral diseases

## INTRODUCTION

In Grenada, as a result of Government incentives to farmers and the increasing demand for eggs and poultry meat, the poultry industry is developing rapidly. Many farmers rear layers and broilers, while a few rear either layers or broilers. Viral and bacterial pathogens cause significant losses in poultry. Arathy *et al*. (2011) reported that out of 143 serum samples from backyard chickens in Grenada, tested by ELIZA, 27 (18.8%) were found to have positive antibody titer for avian influenza virus. In another serological survey Arathy *et al*. (2011) reported presence of antibodies to Infectious bronchitis virus, 54.28% in backyard chickens and 18.02% in broilers. In 2004, a serological survey on backyard chickens (Sharma *et al*., 2006) revealed positivity of 99, 86, 77 and 44%, respectively for Newcastle disease, *Mycoplasma gallisepticum*, infectious bronchitis and *Salmonella enteritidis*. No survey on diseases of commercial chickens in Grenada has been reported. In the present study, we investigated the seroprevalence of important viral diseases in commercial chickens of Grenada.

## MATERIALS AND METHODS

**Ethical approval:** The project was approved by the Institutional Animal Care and Use Committee (IACUC) of St. Georges University.

Birds: source and sampling: The Caribbean island country Grenada divided into six regions (parishes). A total of 226 blood samples from layers (aged between 12 and 18 months) and 233 from broilers (aged between 6 and 7 weeks) were collected from all six parishes of the mainland Grenada. Blood from layers was collected by brachial venipuncture and from broilers at the time of their slaughter. A minimum of 25 blood samples were taken separately from layers and broilers in each parish. Blood was transported in cooler boxes to the Pathology Laboratory of the School of Veterinary Medicine at St. George's University within 2 h of collection. Serum was obtained after routine centrifugation of blood samples. The presence of antibodies against Newcastle disease virus, Avian influenza virus, Avian pneumovirus, Chicken infectious anemia virus and Infectious bursal disease virus in each serum sample was tested by using a commercial ELISA kit (IDEXX Westbrook, Maine, USA), according to manufacturer's instructions.

**Statistical analysis:** Confidence intervals were calculated with the help of the following website, (http://www.mccallumlayton.co.UK/states/confidence intervalCalcProportion.aspx).

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## RESULTS AND DISCUSSION

The seroprevalence of the various viral diseases under study is presented in Table 1.

Newcastle disease (ND) is one of the most important viral diseases affecting poultry and wild birds. Natural or experimental infection has been reported in at least 241 species of birds (Alexander, 2003a). Clinical signs of ND are related to pathotype of the virus strain. Virulent strains (Velogenic) produce respiratory and nervous signs with prostration and death. Mortality is high and in some flocks may reach up to 100%. Infection with less virulent (mesogenic) strains show respiratory signs and drop in egg production in adult birds and mortality around 50%. Low pathogenic (lentogenic) strains do not produce clinical signs in adult birds, however, mild respiratory signs are seen in young birds when there is concurrent infection with other viruses.

ND has been reported from most of the countries of the world. We review here the presence of ND in countries of the Caribbean and South America. Outbreaks of ND have been reported in the Dominican Republic in 2008 caused by highly divergent virulent isolates of NDV (Sean *et al*., 2012) and in Belize where an outbreak of the highly contagious ND was reported in 2009 (Caribbean, 360). ND was diagnosed in Brazil in 2010 (Orsi *et al*., 2010) Venezuela in 2006 (Flu-Trackers, 2006), Chile in 2007 (World Poultry, 2009), Peru in 2009 (Poultry World, 2007) and Argentina (Berinstein *et al*., 1999). Because of the proximity in geographical locations, the presence of ND in South American countries is a threat to the avian population in the Caribbean.

In the present study 47.7% (95% CI, 41.2-54.2%) seropositivity was found in layers and 48.2% (95% CI, 41.8-54.6%) in broilers. This is the first report of the seroprevalence of NDV in commercial chickens in Grenada. Commercial chickens in Grenada are not vaccinated against NDV and in the absence of clinical signs, the study indicates the circulation of a lentogenic strain of NDV in the country. Isolation and identification of NDV is warranted to verify our findings.

Infectious bursal disease (IBD) is an acute highly contagious viral infection in chicken manifested by inflammation and atrophy of the bursa of Fabricius, nephritis and immunosuppression leading to massive economic loss in commercial chickens (Lukert and Saif, 2003). IBDV strains have been classified into 2 distinct serotypes. Serotype 2 includes apathogenic viruses. Serotype 1 includes pathogenic viruses; namely classic strains (CV), very virulent (VV) and variant IBDV.

The disease has world wide distribution including many countries of Latin America and the Caribbean. In Latin America the disease has been reported from Brazil (Di Fabio *et al*., 1999), Venzuela (Banda and Villegas, 2004.) Columbia (Marco, 2007), Uruguay (Hernandez *et al*. 2006) Argentina (Remorini *et al*., 2006). Peru (Marco, 2007) Chile and Bolivia, 2004 (OIE, 2009). IBD is widely distributed in the Caribbean. The OIE bulletin (2009) reported presence of IBDV in Anguilla, Belize, Costa Rica, Cuba, Dominican Republic, Guatemala, Haiti, Honduras and Martinique. To our knowledge there are no reports of IBDV in the East Caribbean Islands including Grenada.

The present study revealed antibodies against IBDV in layers 34.1%, (95% CI, 28.4-40.9%) and broilers 40.3% (95% CI, 34.0-46.6). Chicken and turkey is the natural host of IBDV. Infection is through the fecal oral route and disease is mainly in chickens at young age between 3 and 8 weeks. The absence of acute disease in chickens in Grenada is presumptive of the apathogenic IBDV strain in the country. Since adult birds do not get infected by the fecal oral route, the detection of antibodies in adult layers is unclear. Further studies on the IBDV strains prevalent in Grenada are warranted.

Chicken infectious anemia virus (CIAV) is a member of the Gyrovirus genus of the Circoviridiae family (Pringle, 1999). CIA was first reported in Japan by Yuassa *et al*. (1979) and has since been serologically demonstrated in many countries of the world (Sharma *et al*., 2014). To our knowledge, this is the first report of seroprevalence of CIA from Caribbean region including Grenada.

During present survey the overall seroprevalence of CIA was 92.9% (95% confidence interval, 89.55-96.25%) in layer and 58.3% (95% confidence interval, 51.97-64.63%) in broiler. The higher prevalence in layers compared to that broilers is consistent with previous reports (Ballal *et al*., 2005; Bhatt *et al*., 2011; Owoade *et al*., 2004; Kuyucuoglu *et al*., 2003; Hadimili *et al*., 2008). CIAV is transmitted vertically and horizontally. Clinical disease in vertically transmitted infection occurs during the first 2 weeks of age and is seen as an anemia dermatitis syndrome. Chicks infected horizontally remain subclinical affecting their performance and profitability. Commercial chickens are not vaccinated against CIAV in Grenada. Absence of clinical signs in broilers and multi

Table 1: Seroprevalence of important viral diseases in commercial chickens of Grenada

| | Layers | | Broilers | |
|---|---|---|---|---|
| No. sample tested | 226 | | 233 | |
| Disease agents | No. positive | Percent | No. positive | Percent |
| Newcastle disease virus | 107 | 47.7 | 112 | 48.2 |
| Infectious bursal disease | 77 | 34.7 | 94 | 40.3 |
| Chicken infectious anemia virus | 210 | 92.9 | 136 | 58.3 |
| Avian pneumo-virus | 140 | 61.9 | 74 | 31.7 |
| Avian influenza | 0 | 0.0 | 0 | 0.0 |

age management of layers is indicative of virus circulation within poultry houses and causing horizontal infection of chickens.

Avian peumovirus (APV) is metapneumovirus of the family paramyxoviridae. It is an emerging economically important disease. Serological evidence of APV is now available from many countries of the world (Cook, 1997). A serological survey conducted to detect the presence of APV in Grenada indicated seropositivity of 61.9% (95% CI, 55.6-68.2%) in layers and 31.8% (95% CI, 25.7-37.7%) in broilers (Tiwari *et al*., 2013). Higher prevalence in layers has also been reported by previous researchers (Gough, 2003; Owoade *et al*., 2006). APV is transmitted horizontally and multi age flock management in layers facilitates the transmission. Broilers in Grenada are managed on an "all-in, all-out" system which prevents multi age transmission. At the time of blood collection no clinical signs suggestive of APV were observed in the commercial birds. This finding is in agreement with previous workers who detected antibodies for APV and isolation of this virus in clinically negative birds (Cook *et al*., 1988; Pattison *et al*., 1989; Hafez and Lohren, 1990; Jones *et al*., 1991). Since vaccination for APV is not practiced in Grenada, the presence of antibodies for APV indicates exposure of commercial chickens to this pathogen.

Influenza virus is a member of the family Orthomyxoviridae and includes types A, B, C and Thogoto viruses based on the expression of hemaglutinin and Neuraminidase glycoproteins on the surface of the virus particles. Ducks, geese, swans, gulls, terns and shorebirds are reservoir of all known subtypes of avian influenza virus. In all wild birds, avian influenza virus (AIV) is asymptomatic. However, in poultry and other avian species, AIV produces a range of symptoms from asymptomatic or mild to acute fatal disease (Alexander, 2003b). In acute cases symptoms include watery greenish diarrhea, trembling, edema, cyanosis and hemorrhages of wattles, combs and shanks (Swayne and Halvorson, 2003). Since 2005, AIV has spread from South East Asia to over 60 countries of the world resulting in direct death or slaughter of 250,000,000 poultry (FAO, 2007). These outbreaks emphasize the importance of the need of global surveillance of AIV in natural hosts.

Because of close proximity of the Caribbean islands to South American countries, it will be imperative to review the presence of AIV in the region. In South America there are only few reports of AIV in wild birds. A low pathogenic AI virus (LPAI H7N3) was isolated from a cinnamon teal in Bolivia (Spackman *et al*., 2007). Pareda *et al*. (2010) successfully isolated H13N9 from a gull in Argentina. In Chile, isolations of AIV from gulls was reported during 2007 and 2008 (Dennis, 2010). In Brazil, isolations from 3 species of wild bird, namely a ruddy ternstone, a gull and a sandpiper were reported (Dennis, 2010). In Argentina, antibodies to AIV in wild water birds were detected (Brown *et al*., 2010). In commercial chickens AI has been reported from Columbia and Brazil. LPAI infection in chickens was reported from Brazil during 2006-2007 and in Columbia antibodies to H9N2 were detected in a broiler breeder flock (Dennis, 2010).

Caribbean countries are at high risk of AIV because of large units of the backyard system of poultry production and also due to influence of migratory birds. In the Caribbean, published reports on the detection of AIV are limited. AI was first detected in the Dominican Republic and Haiti in 2007 from live bird market, village poultry and fighting cocks (Dennis, 2010). Between 2006 and 2009, the Caribbean animal health net work surveillance tested wild birds and domestic poultry for AIV from four islands namely Guadeloupe, Martinique, St Lucia and the Dominican Republic. All samples were found to be negative for the AIV matrix gene (Lefrancois *et al*., 2010). So far there is only one report on the isolation and characterization of AIV from wild water fowl in Barbados (Douglas *et al*., 2007). In Grenada, Arathy *et al*. (2011) conducted a virological and serological survey for AIV in backyard chickens, ducks, turkeys, pigeons and guinea fowl. Twenty seven out of 143 (18.8%) serum samples from backyard chickens were positive for AIV antibodies. AIV RNA was not detected in these seropositive chickens. Other species of birds were negative for antibodies and virus isolation.

In the present survey all tested birds (226 layers and 133 broilers) were negative for antibodies to AIV.

The findings in this surveillance study in commercial chickens in Grenada indicate that many important viral diseases are present on the same premises, threatening the productivity of chickens. Our results are in agreement with previous researchers who have also reported similar findings. Biswas *et al*. (2009) reported on the simultaneous presence of AIV, NDV, egg drop syndrome virus, infectious bronchitis virus (IBV) and reovirus in chickens on small holdings in Bangladesh. Similarly, Owoade *et al*. (2006) found seroprevalence of IBV, Avian pneumovirus, infectious laryngotracheitis (ILT) and Avian leukosis virus (ALV) in Nigerian poultry. In their survey too birds were free of antibodies against AIV. Another study conducted in Iran by Shadmanesh and Mokhtari (2013) report the presence of antibodies against ND, AIV, Salmonella, *Mycoplasma gallisepticum* and *M. synoviae* in native hens.

The results of this study will make poultry producers in Grenada aware of disease situation in their commercial birds giving them the opportunity to take appropriate preventive measures. Since the chicken population changes every 2 years, a regular surveillance of important diseases in a new crop of commercial chickens is advised.

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