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# Patho-epidemiological Study of Chronic Respiratory Disease of Poultry in Gonda District of Uttar Pradesh

Rakesh Kumar Gupta<sup>1\*</sup>, Debasish Niyogi<sup>1</sup>, Namita Joshi<sup>2</sup>, Rajesh Kumar Joshi<sup>3</sup>, Jaswant Singh<sup>4</sup>, Abhishek Mishra<sup>1</sup>, Awadhesh Kumar Yadav<sup>1</sup> and Desh Deepak Singh<sup>1</sup> <sup>1</sup>Department of Veterinary Pathology, C.V.Sc. & A.H., ANDUAT, Kumarganj, Ayodhya, Uttar Pradesh-224229, India. <sup>2</sup>Department of Veterinary Public Health and Epidemiology, C.V.Sc. & A.H., ANDUAT, Kumarganj, Ayodhya, Uttar Pradesh-224229, India. <sup>3</sup>Department of Veterinary Microbiology, C.V.Sc. & A.H., ANDUAT, Kumarganj, Ayodhya, Uttar Pradesh-224229, India. <sup>4</sup>Department of Animal Genetics and Breeding, C.V.Sc. & A.H., ANDUAT, Kumarganj, Ayodhya, Uttar Pradesh-224229, India. (Corresponding author: Rakesh Kumar Gupta\*)

(Corresponding author: Rakesh Kumar Gupta\*) (Received 31 December 2020, Accepted 17 March, 2021) (Published by Research Trend, Website: www.researchtrend.net)

ABSTRACT: Chronic respiratory disease is one of the important disease of poultry characterized by manifestation of respiratory signs, decreased egg production, reduced hatchability rates, altered reproductive efficiency of layers and downgrading of carcasses at slaughter. The patho-epidemiological investigation was carried out to know the incidence and mortality patternin poultry affected with chronic respiratory disease in Gonda district of Uttar Pradesh. The naturally dead birds from eight private farms were collected from October, 2018 to March, 2019. The overall incidence was 12.64% with mortality of 9.10%. Grossly, trachea revealed congestion and haemorrhages with catarrhal exudates. Microscopically, trachea revealed thick layer of necrotic material in tracheal lumen with lymphocytic aggregation. Air sacs were covered with caseous exudates and microscopically showed necrotic material with lymphocytic infiltration. Macroscopically, lungs appeared dark red colour showing congestion and haemorrhages. Microscopically, congestion of capillaries and lymphocytic infiltration in interstitial tissue suggestive of interstitial pneumonia. In immunohistochemical examination, *Mycoplasma gallisepticum* antigen was evident as brown coloured staining of air sac epithelial cells.

Keywords: Patho-epidemiology, Gonda, Chronic Respiratory Disease, Uttar Pradesh

# INTRODUCTION

Chronic respiratory disease is one of the important systemic disease with vertical as well as horizontal routes of transmission and thus making the disease economically one of the important disease of poultry. This disease is caused by Mycoplasma species which are pleomorphic and lacks the ability of Gram staining, therefore the characterisation of mycoplasmas based on morphological characteristics is non-effective. Chronic respiratory disease is characterised by coughing, respiratory rales, open mouth breathing, nasal and ocular discharge and air-sacculitis. On post-mortem examination, this disease is characterised by upper and lower respiratory tract lesions which include mild sinusitis, tracheitis and air sacculitis. Nasal passages, infra-orbital sinuses, trachea and bronchi may show presence of catarrhal exudates (McMullin, 2004). Microscopically disease is characterised bv mononuclear cell infiltration and mucosal glandular hyperplasia in the sinus and trachea (Salami, 1994). membranes Affected mucous are thickened, hyperplastic and necrotic. The mucosal lamina propria contains focal areas of lymphoid hypoplasia and germinal centre formation. In the lungs, interstitial pneumonia and lymphoid follicular reactions may be seen (Charlton *et al.*, 1996; Gupta *et al.*, 2020). Immunohistochemistry has been proved as an excellent tool for effective detection of mycoplasma antigens in many animals including avian species (Nunoya *et al.*, 1997; Radi *et al.*, 2000; Yilmaz *et al.*, 2011). The main aim of this study was to diagnose the Chronic Respiratory Disease by immunohistochemistry so that disease could be reduced or prevented and more income can be generated by the poultry farmers.

## MATERIALS AND METHODS

## A. Study design

For the present study, poultry birds affected with chronic respiratory disease were screened in eight different private and commercial farms (broiler and layer) located in Gonda district of Uttar Pradesh for six months starting from October, 2018 to March, 2019. Two private farms from each tehsil of Gonda district were selected for the above purpose on the basis of stratified random sampling. These farms were visited

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during study period to screen disease on the basis of clinical signs and symptoms like respiratory rales, open mouth breathing, ocular and nasal discharge, marked depression and weakness, decreased feed intake, feed conversion, egg production and hatchability.

#### B. Epidemiological Study

The epidemiological study includes incidence rate, mortality rate and case fatality rate.

# C. Post-mortem examination

The naturally dead birds from the above different private farms were collected and brought to the Veterinary Pathology Laboratory, ANDUAT, Ayodhya for the post-mortem examination. The post-mortem examinations were conducted thoroughly and carefully for any gross lesions in the trachea, air sacs and lungs. Post mortem examination was also conducted at the farms.

## D. Histopathological and Immunohistochemical study

The samples were collected in 10% neutral buffered formalin for further histopathological and immunohistochemical studies. The representative tissue pieces from samples were fixed in 10% formal saline and were processed for paraffin block and sectioning into 3-5 mm thickness by microtome machine and were stained with Haematoxylin and Eosin (H&E) stain for examination histopathological (Luna, 1968). Immunohistochemical study was carried out as per the procedure of Brar et al., (2017) using primary antibody, chicken polyclonal to *Mycoplasma gallisepticum* (Abcam, UK).

## **RESULTS AND DISCUSSION**

Out of total 34400 birds at the eight farms, 4351 were found to be naturally affected with Chronic Respiratory Disease. The overall incidence of the Chronic Respiratory Disease being 12.64% which corroborated with the findings of Sultana et al., (2012), Ahmad et al., (2009) and Hasan et al., (2016) who also reported the incidence of the disease in birds ranging from 11.81 to 14.70% but was in contrast with the findings of Abbas et al., (2015) and Islam et al., (2003) who reported incidence as 3% and 5.32% respectively. The incidence varies from 3.75 to 16.12% in different farms. Such difference might be due to variations in nutritional status, husbandry practices from region to region and other superimposed infections like E. coli infection, IB, IBD, ILT etc. In the present study, the overall mortality being 9.10% simulated the findings of Nunoya et al., (1995) and Barua et al., (2006) who reported mortality ranging from 1.0 to 10% and 10% respectively but was in contrast with the findings of Shrivastav (1990), Chary et al., (1991) and Jordon and Pattison (1996) as 0.82%, 0.44% and 1 to 2 % respectively. The mortality varies from 0.65 to 11.92%. This was probably due to variation of geoclimatic conditions, nutritional, toxic patho-physiological conditions. Detailed and epidemiological information about the farm strength, age of birds, morbidity, mortality and case fatality rate is given in Table 1.

 Table 1: CRD diagnosed in eight farms under study along with morbidity, mortality patterns and age of the affection.

Farm No.	Total number of birds	Number affected	Age of infection	Number of Death	Morbidity (%)	Mortality (%)	Case fatality rate (%)
1	4200	434	30-38 days	290	10.33	6.90	66.82
2	7000	1002	23-28 weeks	781	14.31	11.15	77.94
3	8000	1290	25-32 weeks	954	16.12	11.92	73.95
4	3000	330	26-35 days	233	11.00	7.76	70.60
5	4500	640	27-42 days	442	14.22	9.82	69.06
6	2200	218	28-36 days	129	9.90	5.86	59.17
7	3500	362	22-29 days	289	10.34	8.25	79.83
8	2000	75	11-14 days	13	3.75	0.65	17.33
Total	34400	4351	-	3131	12.64	9.10	71.96

Mild to moderate congestion and haemorrhageswere observed in the trachea (Fig. 1) Thick and purulent exudates were adherent to tracheal wall. Histopathology of trachea revealed a thick layer of necrotic material in the tracheal lumen with the loss of epithelium and cilia and lymphocytic aggregation (Fig. 2). Catarrhal inflammation was found in tracheas along with epithelial degeneration, destruction of cilia and desquamation. The gross and microscopic lesions observed in the present study were similar with the findings of Islam *et al.*, (2011), Vitula *et al.*, (2011), Brar *et al.*, (2017) and Karthik *et al.*, (2018).

The air sac become thickened and covered with caseous exudates and classical cloudiness was seen (Fig. 3). Histologically air sac showed necrotic material adhered in the air sac wall along with lymphocytic infiltration. Air sac were moderately to severely thickened with severe lymphocytic infiltrates. The gross and microscopic lesions observed in the present study are almost similar to those recorded by Gharaibeh and Hailat (2011), Islam *et al.*, (2011), Vitula *et al.*, (2011), Brar *et al.*, (2017) and Karthik *et al.*, (2018).



Fig. 1. Congestion and haemorrhage in trachea of affected bird.



**Fig. 2.** Thick layer of necrotic material in the tracheal lumen with the loss of epithelium and cilia along with lymphocytic aggregation. H & E X 100.



Fig. 3. Air sacculitis with cloudy appearance in the air sac of affected bird with CRD.

Grossly, Lungs were consolidated, haemorrhagic and frothy exudates came out from cut surface (Fig. 4). Microscopically, lung section revealed congestion and haemorrhages, lymphomononuclear cell infiltrates in interstitial area besides necrotic and denuded epithelial cells which are suggestive of interstitial pneumonia (Fig. 5). The gross and histopathological observations of lung sections corroborated the earlier reports of Islam *et al.*, (2011), Vitula *et al.*, (2011), Thilagavathi *et al.*, (2016), Brar *et al.*, (2017), Karthik *et al.*, (2018), Ley (2018) and Gupta *et al.*, (2020).



Fig. 4. Consolidated and haemorrhagic lung of affected bird.



**Fig. 5.** Congestion, haemorrhages and lymphocytic infiltration in interstitial area of lung (interstitial pneumonia). H & E X 100.

In the present study, the immunohistochemical studies performed on the suspected cases of chronic respiratory disease as per the gross and histopathological findings. *Mycoplasma gallisepticum* antigen was evident as intense brown coloured staining of air sac epithelial cells (Fig. 6). Similar findings were observed in other studies of Yilmaz *et al.*, (2011), Gharaibeh and Hailat (2011), Yilmaz and Timurkaan (2011), Casagrande *et al.*, (2014), Brar *et al.*, (2017) and Gupta *et al.*, (2020) where *Mycoplasma gallisepticum* antigen were detected in epithelial cells of air sacs, primary bronchus as well as cilia of intrapulmonary primary bronchus epithelial cells in infected chickens.

The present study reports chronic respiratory disease in poultry population in Gonda district of Uttar Pradesh, India and explores the previous limited epidemiological data of this disease. Further, diagnostic tests like PCR, gene sequencing, phylogenetic analysis etc. are required for the diagnosis of this disease. The diagnosis performed by gross and histopathology should also involve molecular detection tools to assess the extent of

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the lesions and epidemiology. The explorative surveillance and epidemiological studies in other parts of Uttar Pradesh are required to find out the real effect of this economically important disease of poultry.



**Fig. 6.** *Mycoplasma gallisepticum* positive brown colour (antigen-antibody reaction) in the desquamated epithelial cells and cytoplasm of inflammatory cells of air sacs. IHC X 200.

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