

HJARRE'S DISEASE IN CHICKENS: CLINICAL, PATHOLOGICAL, MICROBIOLOGICAL AND THERAPEUTIC FINDINGS

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ABSTRACT

Hjarre's disease (Coligranuloma) in commercial chickens was investigated and detected in 5 layer flocks out of 47 outbreaks based on clinical, pathological, microbiological and therapeutical findings. The investigation areas were at Dinajpur and Nilphamari districts, but the laboratory examinations were conducted at Dinajpur Government Veterinary College, Dinajpur, Bangladesh. The flocks had 441, 212, 690, 5400 and 735 birds aged between 26 to 64 weeks and reared in cages. The clinical signs of the affected birds varied markedly from farm to farm and diarrhoea, depression, soiled vent, reduces egg production, loss of body condition and death were recorded. The morbidity rate was around 100%, but the mortality rate was 3-9%. At necropsy the birds showed characteristics nodular lesions on the serosal surface of the intestine, uterus, mesentery and in liver. The affected organs were processed for the bacteriological examination and the organisms were isolated and identified without typification. Gram stained impression smears from the necrotic areas of the nodules revealed gram negative organisms which were isolated by culturing using different agar media and identified by their colony characteristics including biochemical reaction in differential media. No acid fast organisms found on acid fast staining of the impression smears. The collected organs were processed and stained for the histopathological study, where granulomatous inflammation and the characteristic caseation necrosis were recorded. The flocks were treated individually either with colistin sulphate, or fluoroquinolones, or oxytetracycline considering the apparently presence of concurrent infection. The present findings indicated that the poultry is sensitive to colistin sulphate.

Key words: Hjarre's disease, *E. coli*, clinico-pathology, therapy, chickens

INTRODUCTION

Avian colibacillosis is an infectious disease of birds in which *Escherichia coli* is the primary or secondary pathogens. Infection includes airsacculitis, cellulitis, omphalitis, peritonitis, pericarditis, salpingitis, synovitis and Hjarre's disease (coligranuloma) is caused by *E. coli* (Charlton, 2000, Calnek *et al.*, 1997). The organisms are the normal inhabitants of the intestine of the birds and outbreaks occur in poultry raised below standard sanitation, poor environmental condition, or during the course of respiratory or immunosuppressive diseases like mycoplasmosis, Newcastle disease, infectious bronchitis, infectious laryngotracheitis or others (Charlton, 2000; Fraser *et al.*, 1998). Colibacillosis is common throughout the world including Bangladesh. But coligranuloma is one of the important forms of colibacillosis and usually sporadically found in adult birds (Chauhan, 2003). The clinical signs are nonspecific (Vegad and Katiyar, 2003; Charlton, 2000), but often appeared with diarrhoea, soiling of cloaca with semisolid cheesy materials, and death (Chauhan, 2003). Large numbers of grayish-whitish nodules are commonly found on the serosal surface of the intestine, mesentery, and in liver. Granulomatous inflammation with caseation necrosis was recorded on histological examination. The organisms isolated and identified on the basis of cultural characteristics and biochemical reactions were *E. coli*. The organisms give good response to colistin sulphate, oxytetracycline, fluoroquinolones, neomycin (Charlton, 2000). The aim of the study was to diagnose the disease based on clinical, pathological and microbiological findings as well as to evaluate the response to certain commercially available antibiotic therapy at field level without conducting any drug sensitivity test.

MATERIALS AND METHODS

Experimental chickens/Case history

Hjarre's disease (Coligranuloma) in commercial chickens was investigated at Dinajpur and Nilphamari districts, but the laboratory examinations were conducted at Dinajpur Government Veterinary College, Dinajpur, Bangladesh. Forty seven outbreaks in layer aged between 26 to 64 weeks reared in cages were investigated. The detailed flock history including types of birds, population of birds/flock, rearing system, age of birds, morbidity, mortality as well as the number of birds examined was presented in the Table 1. The clinical signs were recorded during the physical visit of the affected farms and the farmer's complaints were also considered. Five out of 47 outbreaks were investigated extensively as they were found to be infected with *E. coli*.

Table 1. Clinical history of the different flocks affected by Hjarre's disease

No. of flocks	Flock size	Age of birds (weeks)	Morbidity (%)	Mortality (%)	No. of birds examined at necropsy
Flock 1	441	36	Around 100	4	3 - 5 birds/flock
Flock 2	212	47	As above	7	As above
Flock 3	690	33	As above	5	As above
Flock 4	5400	26	As above	3	As above
Flock 5	735	64	As above	9	As above

Pathological examination

The bird was examined systematically and the postmortem changes observed were recorded during necropsy (Charlton, 2000). The representative tissues were collected and preserved at 10% formalin solution and subsequently processed, embedded with paraffin, sectioned and stained with haematoxylin and eosin for histopathological examination (Luna, 1968). The slides were examined under low and high power objectives and the lesions were recorded.

Microbiological examination

The tissue samples were collected and brought to the bacteriology laboratory with necessary precautions. Impression smears taken from the collected samples was stained with Gram's stain (McLeod *et al.*, 1981) to demonstrate the bacteria. Acid fast staining was also carried out to differentiate the organisms from the tuberculous bacilli following a recommended procedure (McLeod *et al.*, 1981).

Isolation of the bacteria in culture from the collected tissue samples were made by standard routine laboratory methods (Anon., 1984) by using different media (Nutrient agar, Blood agar, MacConkey agar, Eosin Methylene Blue or EMB agar, Salmonella-Shigella or SS agar) and their colony and staining characteristics including cellular morphology were noted.

Biochemical tests including catalase, coagulase, indol production, methyl red tests were conducted. The results of the tests were recorded. Identification of the bacteria was determined on their cellular morphology and arrangements, staining and colony characteristics as well as biochemical reactions.

Therapeutic trial

Colistin sulphate (Colexin WS, Advance Animal Science, Bangladesh) @ 1 g per litre of drinking water daily for 5 days, oxytetracycline (Renamycin Powder, Renata Animal Health, Bangladesh) @ 1 g per 2 litre of drinking water daily for 5 days and fluoroquinolones [Flumequine 10% Powder, Arifs (Bangladesh) Limited] @ 1 g per litre of drinking water daily for 5 days were utilized for the treatment of coligranuloma in different farms based on the necropsy findings only. The antibiotics were selected mostly considering the apparently presence or absence of concurrent infections during necropsy findings. The antibiotic course was maintained up to 5 days as per recommendation. Drug sensitivity test for the selection of antibiotic was not performed. Vinegar @ 10 ml per litre of water daily for 5 days was used along with antibiotic as acidifier. The suggestion for improving the sanitation in poultry houses and the management practices to avoid any stress was given along with the treatment. The response to treatment was recorded.

RESULTS AND DISCUSSION

Hjarre's disease (Coligranuloma) in commercial chickens was investigated based on clinical, pathological, microbiological and therapeutic findings. The disease was diagnosed on the basis of clinical signs, necropsy findings, histopathological examinations, the isolation and identification of organisms by bacteriological examination, the positive therapeutic response of the affected flocks to commercially available antibiotics. Coligranuloma is a granulomatous bacterial infection of chickens. The disease appeared sporadically and commonly seen in adult fowls. The detailed clinical history of the affected flocks was summarized in Table 1. The morbidity of all affected flocks was around 100%, but mortality was varied from farm to farm ranging from 3-9% (Table 1). The clinical signs observed during the farm visit varied from farm to farm but found to correlate with previous findings (Charlton, 2000; Vegad and Katiyar, 2003). Diarrhoea, depression, soiling of cloaca with semisolid cheesy material, and loss of condition (Chauhan, 2003; Vegad and Katiyar, 2003) were recorded.

On postmortem examination, large number of grayish-whitish nodules of varying degrees in sizes found projecting from the serous surface throughout the length of the intestine, but fewer in the descending portion (Fig. 1 - 4) (Sastry, 1983; Chauhan, 2003). The nodules were discretely distributed in the mesentery and liver, but not found in spleen (Charlton, 2000). Granulomatous nodules were similarly found on the uterine surface. The characteristic gross lesions (singly distribution of the nodules on the serosal surface of intestine, uterus and in liver distinctly differentiated the coligranuloma from avian tuberculosis (Sastry, 1983).

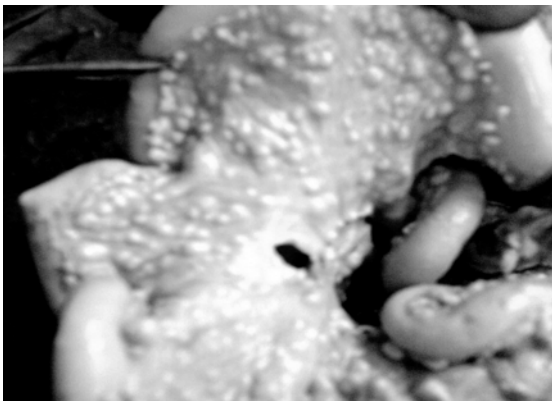


Fig. 1. Discrete nodules are present over the serosa of intestine.

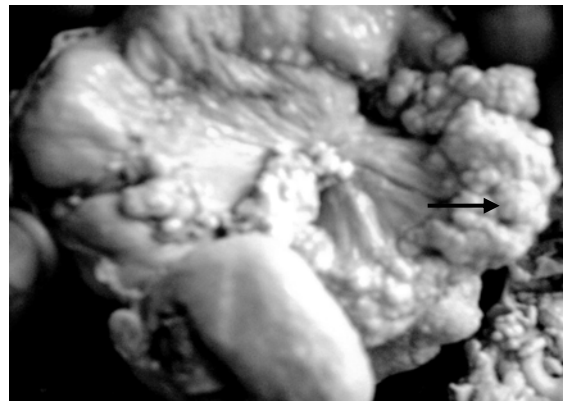


Fig. 2. Discrete nodules are present over the serosa of uterus (arrow indicates the aggregated nodules).

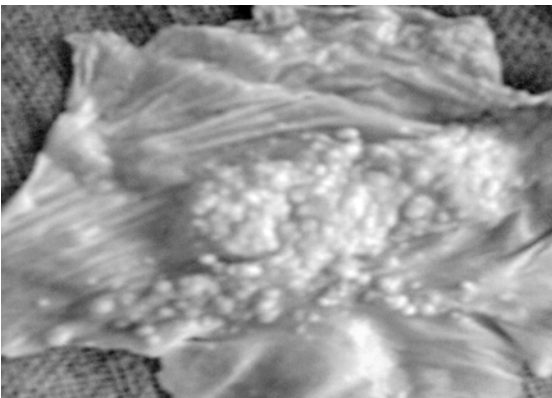


Fig. 3. Discrete nodules are present over the serosa.

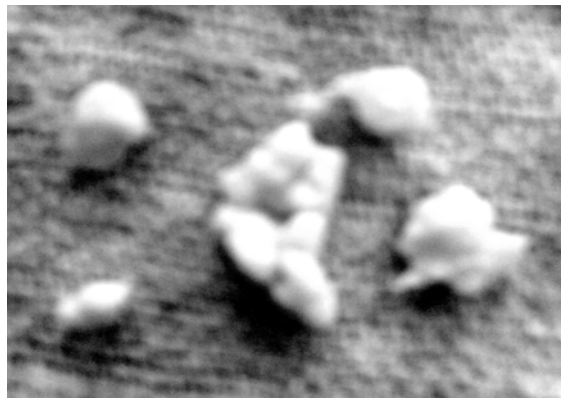


Fig. 4. Nodules are isolated from the lesions.

Cultural, staining and biochemical characteristics

A presumptive diagnosis was made on the basis of examination of Gram stained impression smears of dissected nodules (McLeod *et al.*, 1981; Anon., 1984), where large number of gram negative rod shaped organisms were found, but no acid fast organisms were observed on examination of impression smears of dissected nodules with acid fast staining. The organisms were readily isolated from the lesions and identified by aerobic incubation overnight at 37°C directly on different agar media, where smooth, large, grayish-white, circular colonies on blood agar; rose pink colored colonies on MacConkey agar and the characteristic green sheen on EMB agar (Levinson and Jawetz, 2000) were recorded (Table 2). The organisms produced acid and gas on fermentation.

Table 2. Colony characteristics, staining reaction and biochemical properties of the organisms isolated from the granulomatous tissues

Nutrient agar	Smooth white to grayish white colony
Blood agar	Produced haemolysis
MacConkey agar	Rose pink lactose fermented colony
Eosin Methylene Blue (EMB) agar	Yellow green metallic sheen
Salmonella-Shigella (SS) agar	Pink colour colony
Shape	Short plump rod
Arrangement	Singly, paired, short chained
Gram's staining	Negative
Acid fast staining	Negative
Basic sugar fermentation	Positive with production of acid and gas
Catalase test	Positive
Coagulase test	Negative
Indol production test	Positive
Methyl red test	Positive
Isolated organism	<i>Escherichia coli</i>

Histopathological observation

Centrally purple colored granular mass surrounded by macrophages, lymphocytes, epitheloid cells and a few giant cells were found. Desquamated intestinal epithelium in the affected portion was recorded (Sastry, 1983) on histopathological examination.

Antibiotic sensitivity

The flocks were treated by the oral administration of colistin sulphate, fluoroquinolones, where apparently concurrent infection was not suspected, and oxytetracycline, where apparently concurrent infection was suspected. Better response to colistin sulphate was recorded. Along with the course of antibiotic therapy strict hygienic measures was also emphasized. The disease in adult fowls was diagnosed basis on the characteristic gross and histopathological lesions, identification of gram negative bacilli, colony characteristics, the positive biochemical reactions and good response to treatment.

However, investigation of Hjarre's disease in commercial chickens based on the findings as stated above will certainly help in proper diagnosis of the disease which is sporadically found in Bangladesh and causes a considerable economical loss to the poultry sectors. So, this study will also help to dictate specific medication as well as to take proper prevention and control strategies.

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