Journal of Animal Research: v.13 n.02, p. 265-268. April 2023

**DOI:** 10.30954/2277-940X.02.2023.14

# Colibacillosis Outbreak in One-Week Old Karaknath Chicks at an Organized Poultry Farm in Meerut, Uttar Pradesh

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**Received:** 27 Jan., 2023 **Revised:** 08 March, 2023 **Accepted:** 14 March, 2023

**ABSTRACT**

*Escherichia coli* is one of the most important pathogens in poultry farms around the world, causing colibacillosis and early chick mortality. Outbreaks cause heavy economic losses as primary pathogen and comoribund with other diseases. The report presents an outbreak of colibacillosis with mortality of 12.50% in one-week old Karaknath chicks at an organized poultry farm in Meerut. The cases were diagnosed on the basis of history, clinical findings, postmortem investigation and laboratory examination. The ailing chicks were weak and dejected, with stunted growth, respiratory discomfort and vent pasting. Postmortem examination revealed polyserositis with fibrinous hepatitis, pericarditis, peritonitis, air sacculitis and omphalitis. The *E. coli* isolates were obtained from the infected tissues of dead chicks and cloacal swabs of sick birds using bacteriological and biochemical techniques. The isolates were resistant to amoxicillin/clavulanic acid, ampicillin, cefuroxime, tetracycline, norfloxacin and enrofloxacin, but showed sensitivity to ampicillin/sulbactum, amikacin and gentamicin. The apparently healthy and ailing chicks of the farm were treated with gentamicin @ 5-10 mg/kg body weight orally for 5 days. Multivitamin @ 5 g/L was given in drinking water for 3 days to improve the immunity. Treatment resulted in clinical recovery of the chicks and checked further mortality at the farm.

**HIGHLIGHTS**

- Outbreak of Colibacillosis in one week old Karaknath chicks.
- Carcass of chicks revealed polyserositis with marked omphalitis.
- Gentamicin was antibiotic of choice for treating ailing chicks resulting in recovery.

**Keywords:** *Escherichia coli*, colibacillosis, Karaknath chicks, antibiotic sensitivity

One of the most prevalent infectious diseases in poultry of all ages is colibacillosis, which often affects young chicks up to three weeks old (Kabir, 2010; Singh *et al.,* 2018). Due to its association with various disease conditions as either a primary or secondary infection, it is one of the main causes of financial losses for the poultry industry globally (Koutsianos *et al.,* 2020; Xing *et al*., 2021).

Any localised or systemic infection brought on entirely or in part by Avian Pathogenic *Escherichia coli* (APEC) is referred to as avian colibacillosis. This includes colisepticaemia, hemorrhagic septicemia, coligranuloma (Hjarre's disease), swollen head syndrome, venereal colibacillosis, coliform cellulitis, pericarditis, perihepatitis, airsacculitis, peritonitis, salpingitis, orchitis, osteomyelitis/synovitis, panophthalmitis, omphalitis/yolk sac infection and enteritis (Panth, 2019).

*E. coli* is a member of the family *Enterobacteriaceae* and is a gram-negative, non-acid-fast, non-spore-forming

**How to cite this article:** Katoch, S., Singh, R., Verma, H., John, J.K. and Jaiswal, V. (2023). Colibacillosis Outbreak in One-Week Old Karaknath Chicks at an Organized Poultry Farm in Meerut, Uttar Pradesh. *J. Anim. Res.,* **13**(02): 265-268.

**Source of Support:** None; **Conflict of Interest:** None

bacillus that is typically 3 × 0.6 mm in size (Nolan *et al.,* 2013). It is usually motile, often fimbriated, with peritrichous flagella. *E. coli* serotypes are distinguished by their somatic (O), flagellar (H), and occasionally capsular (K) antigens.

For colibacillosis, the incubation period is three to five days. Infection is transmitted vertically by transovarian route and horizontally by oral or inhalation routes, and via shell membranes through faeces, contaminated feed, water and fomites (Kabir, 2010). Mortality ranges from 5-20% while morbidity varies.

In this report, we investigated the outbreak of colibacillosis in an organised poultry farm in Meerut district of western Uttar Pradesh. In order to aid in the future control of the disease, the isolates were examined to ascertain their susceptibility to antimicrobial drugs. The report also emphasises the step-by-step process utilised to arrive at a diagnosis and course of treatment. The present report appears to be the first documentation of outbreak of colibacillosis in one week old Karaknath chicks in an organised poultry farm of the western Uttar Pradesh.

## MATERIALS AND METHODS

### Case history

An organised poultry farm in the Meerut district of western Uttar Pradesh reported mortality in Karaknath chicks that were one week old during the rainy season. When the case was reported and a thorough post mortem examination was carried out, there had been a total mortality of 25 chicks (12.50%) from a stock of 200 chicks. The chicks were raised intensively on the floor in the brooders. History further revealed that the chicks had an intraocular vaccination against Newcastle disease at a day old age. The chicks have not yet received prophylactic antiparasitic medication. The affected chicks were reportedly depressed, weak and dejected, with stunted growth, showing respiratory discomfort with vent pasting. Individual birds who were severely affected exhibited a lack of activity, a tendency to withdraw themselves from the group, closed eyes, a stooped posture, and drooping head, neck and wings. Even when Amoxy/Clav (Amoxicillin/clavulanic acid) antibiotic treatment was administered to the entire flock for three days at a dosage of 500 mg per L of drinking water prior to the case being reported for investigation, there was no reduction in mortality rate.

### Sample collection and post-mortem examination

A total of 12 deceased chicks were submitted to department of Veterinary Microbiology for the examination. The detailed post-mortem examination was performed on the presented carcases and the gross lesions were noted. The tissues from heart, liver, lung, spleen and intestine were aseptically collected from the dead chicks and were processed for bacterial isolation, identification and antimicrobial susceptibility testing. A total of 8 cloacal swabs were also collected aseptically from the depressed chicks with vent pasting, after thoroughly cleaning the vent area with normal saline and wiping it with 70% ethanol swab and were subjected for bacterial examination.

### Bacterial isolation and identification

The tissue samples and cloacal swabs collected were inoculated on 5% defibrinated sheep Blood agar (BA), MacConkey Lactose agar (MLA) and Eosin Methylene blue agar (EMB) and plates were incubated aerobically at 37°C for 24 hours. After 24 hours of incubation the colonies were examined for their cultural and morphological characteristics. The organism was stained with Gram's staining method (Oh *et al.,* 2011). The isolates were identified using primary and secondary biochemical tests (Quinn *et al.,* 2011).

### Antimicrobial susceptibility testing

The *in vitro* antimicrobial susceptibility test of the isolates was conducted on Mueller-Hinton Agar (MHA) as per the disk diffusion method (Jorgensen and Turnidge, 2007), using amikacin, amoxicillin/clavulanic acid, ampicillin, cefuroxime, enrofloxacin, gentamicin and tetracycline. Overnight grown bacterial colonies were suspended in 5 ml nutrient broth and incubated at 37°C to obtain the turbidity equivalent to a 0.5 McFarland standard. 100 µl of bacterial suspension was spread over the MHA plate and antibiotic discs were placed aseptically on the surface of inoculated medium. Results were recorded after 18 hours of incubation at 37°C. The efficacy of antibiotics was determined by measuring the diameter of zones of inhibition around the discs and interpretation

followed criteria recommended by Committee for Clinical Laboratory Standards (2020).

## RESULTS AND DISCUSSION

### Gross Pathology

The gross lesion that was seen most frequently was polyserositis, which included fibrinous hepatitis, pericarditis, peritonitis, air sacculitis, and marked omphalitis. The air sacs had adherent caseous deposits and were opaque, thick and white in colour. In all the necropsied chicks, swollen naval and a congestive yolk sac with aberrant material were seen. The gross lesions seen in the investigation are consistent with the cases of Colibacillosis reported previously by Yousseff *et al.* (2008), Kabir (2010), Tonu *et al.* (2011), Daud *et al.* (2014), Parwez *et al.* (2015) and Singh *et al.* (2018). The virulence factors of *E. coli* have a close correlation with the pathogenicity of the infection (Kaper *et al.,* 2004; Pakbin *et al.,* 2021). According to Tonu *et al.* (2011), *E. coli* strains were classified as pathogenic or non-pathogenic based on their propensity to cause disease as well as the presence of different virulence factors or a combination of them.

### Bacterial isolation and identification

The cultures from the tissues and cloacal swabs on 5% Sheep Blood Agar yielded pure growth of one type of colonies. The colonies were mucoid, with white to greyish convex appearance. The subculture of these colonies on MLA revealed lactose fermenting pink coloured colonies and on EMB metallic sheen colonies were observed. On Gram staining, pink coloured rod shaped Gram negative bacteria were observed. Similar morphological, cultural and staining characteristics of *E. coli* were described by other researchers (Jakaria *et al.,* 2012). The isolates were further confirmed by biochemical tests and all were motile, positive to Catalase test, Indole test, Methyl red test, Triple sugar iron agar test and Nitrate reduction test and negative to Oxidase test, Voges Proskaur test, Citrate utilization test, Urease test and $H_2S$ production which is in agreement with earlier findings of Quinn *et al.* 2011. These findings corroborated the unequivocal presence of colibacillosis at a farm as reported by Nolan *et al.* (2013) and Daud *et al.* (2014), and allowed for the identification of the bacterial isolates as *Escherichia coli*.

### Antimicrobial susceptibility testing

The isolates were sensitive to ampicillin/sulbactum, amikacin and gentamicin but they were resistant to amoxicillin/clavulanic acid, ampicillin, cefuroxime, tetracycline, norfloxacin and enrofloxacin. These findings are similar to the earlier studies of Omer *et al.* 2010 where high resistance of *E. coli* isolates to ciprofloxacin, chloramphenicol, piperacillin/tazobactam, ceftizoxime, cefotaxime, pefloxacin, tetracycline and ampicillin/sulbactam and sensitivity to amikacin, gentamicin, co-trimoxazole and ofloxacin was reported. In the present report, the isolates demonstrated high resistance to most commonly used antibiotics. This is most likely due to increased use of antibiotics as feed additives for growth promotion or prevention of diseases as reported by Bejar *et al.* 2021 and Vidovic & Vidovic, 2020. Inappropriate use of antibiotics (Ventola, 2015), the transmission of resistance among different bacteria (Wintersdroff, 2016), overcrowding, and inadequate sanitation are additional factors that contribute to antimicrobial resistance. The antimicrobial susceptibility test in the report revealed resistance of isolates to Amoxy/Clav which explains its ineffectiveness in controlling the morbidity and mortality in chicks prior to the investigation.

### Treatment

Gentamicin was administered in drinking water for 5 days based on the findings of an antimicrobial susceptibility test at a dose of 5 to 10 mg/kg body weight. To boost the immunity of chicks, multivitamin powder @ 5 g/L was administered in drinking water for 3 days. Treatment resulted in clinical recovery of the chicks and checked further mortality at the farm.

## CONCLUSION

This report confirmed the outbreak of colibacillosis in one-week old Karaknath chicks in an organised farm in Meerut. According to the findings of this report, timely identification based on post-mortem examination, bacterial culture and antibiotic sensitivity testing is essential for treating and effectively managing colibacillosis in poultry farms. To prevent this economically significant disease in chicken, it is imperative to implement a regular monitoring programme to track antimicrobial resistance

in pathogenic bacteria, a good management system and stringent biosecurity measures.

## ACKNOWLEDGEMENTS

The authors thank the Hon'ble Vice-Chancellor & Dean, College of Veterinary & Animal Sciences, Sardar Vallabhbhai Patel University of Agriculture & Technology, Meerut for providing necessary facilities of work.

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