

## Original Research Article

# Incidence of spontaneous *E. coli* Infection in Broiler Chickens in Faizabad and Sultanpur Districts of Uttar Pradesh

G. K. Singh<sup>1\*</sup>, D. Niyogi<sup>1\*</sup>, K. K. Tripathi<sup>1</sup>, R. K. Joshi<sup>2</sup>, S. V. Singh<sup>3</sup> and P. K. Choudhary<sup>4</sup>

<sup>1</sup>Department of Veterinary Pathology, N. D. University of Agriculture and Technology, Kumarganj, Faizabad-224 229, India

<sup>2</sup>Department of Veterinary Microbiology, NDUAT, Kumarganj, India

<sup>3</sup>Department of Veterinary Medicine, NDUAT, Kumarganj, India

<sup>4</sup>Department of Veterinary Physiology and Biochemistry, NDUAT, Kumarganj, India

\*Corresponding author

## ABSTRACT

A study was undertaken round the year from May 2015 to April 2016 to establish the incidence of spontaneous *E. coli* infection in broiler chickens in Faizabad and Sultanpur districts of Uttar Pradesh. For this study a total number of 178 naturally dead broiler chickens from different private farms of Faizabad district and 151 naturally dead broiler chickens from Sultanpur district were collected throughout the year. The birds were of different age group and of both sexes. After post mortem examination, the grossly suspected samples were kept for further bacteriological study. In Faizabad district overall incidence of colibacillosis was 53.37% (that is 95 samples were positive out of 178 samples). In Sultanpur district, 29.13% birds (that is out of 151 samples, 44 samples were positive) for *E. coli* infection. Postmortem examination was conducted for all birds (178+151) and different internal organ were examined carefully for presence of gross lesions. The gross change of liver was found as enlarged, congested and covered with thick yellow/white serofibrinous covering. In intestine, hemorrhages, congestion and edematous swelling were noted. Heart was also covered with thick yellow/white serofibrinous covering. Among tested 178 and 151 samples from Faizabad and Sultanpur, 95 (53.37%) and 44 (29.13%) were found to be suggestive of *E. coli* giving typical large (2-3mm) lactose fermenting pink coloured colonies on MacConkey's agar plate. They again produced characteristic 'metallic sheen' on EMB agar. On Gram staining during morphological study, they produced pink coloured, rod shaped ones (that is gram negative) with characteristic arrangement. All these isolates (from district Faizabad) showed typical biochemical reaction which were positive to Indole test, M.R. test, T.S.I. agar test, nitrate reduction test and negative to VP test and H<sub>2</sub>S production test. Among 95 *E. coli* isolates 97.89% were negative to citrate utilization test and 96.84% to urase activity test. Out of 95 isolates of *E. coli*, 91 were motile. They all fermented D-glucose, lactose, manitol. Among 95 isolates of *E. coli* 68 (71.57%) isolates, 71(74.73%) isolates and 57 (60.00%) isolates had shown fermentation of sucrose, dulcitol and salicin respectively. Only two isolate fermented adonitol. The isolates from the farms of Sultanpur also showed the typical reaction. They were positive to Indole test, MR test, TSI agar test, nitrate reduction test. They had shown 100% motility and negative to VP and H<sub>2</sub>S production test. They had shown 88.63 % (39 birds) negative result to citrate utilization and urase activity test. They all fermented D-glucose and lactose. Among the 44 isolates of *E. coli*, 72.72% (32) isolates, 86.36% (38) isolates, 86.36% (38) isolates, 25% (11) isolates and 88.63% (39) isolates had shown fermentation of sucrose, mannitol, dulcitol, adonitol and salicin respectively.

## Keywords

Broiler, chicken,  
*E. coli*, incidence

## Introduction

With all the virtues of poultry rearing, the industry is now in threat of a number of infectious, contagious and emerging diseases causing substantial economic losses. There are so many pathogens which are responsible for mortality and morbidity in broiler birds. The bacterial count in poultry housing systems is high in comparison to those of pig and cattle. Little is known about the bacteria present in the poultry environment such as in poultry litter and air of poultry house (Saleh *et al.*, 2003). Bacteria present in poultry environment may enter into the flock to produce disease. There are also reports indicating that poultry feed and water may act as a source for various infectious diseases (Rahman *et al.*, 1999). In addition, poultry itself may act as a reservoir or source of infectious agent for other healthy birds. Therefore, it is important to know the prevalence and distribution of different bacterial flora in poultry and its environment as many of them may be potential pathogen for poultry. Such information is also required to take necessary actions for the prevention and control of diseases caused by bacterial pathogens.

Avian colibacillosis is one of the most common infectious diseases seen in poultry of all age group and usually found in young chick of up-to three weeks of age. It occurs in all poultry as well as in other birds and mammals. Colibacillosis is an important cause of mortality in poultry of younger age than older one (Kabir, 2010). Economically the disease is very important as it causes heavy mortality producing heavy economic loss. Avian pathogenic *E. coli* (APEC) strains belong to the ExPEC group is a major pathogen responsible for morbidity and mortality in chickens (Ashraf *et al.*, 2014). Further it suppresses immune system

of the chicks and makes them to get infection easily. Zoonotically, the disease is very important as poultry meat is richest source of protein to human population of world (Ewers *et al.*, 2003). Amongst poultry diseases transmissible to human being, avian colibacillosis is of great concern. Keeping in view the immense importance of this disease in broiler industry, the present study was undertaken to investigate the incidence of *E. coli* infection in broiler chicken in two districts (Faizabad and Sultanpur) of Eastern Uttar Pradesh.

## Materials and Methods

For the present study, a total number of 178 naturally dead broiler chickens from different private farms of Faizabad district and 151 naturally dead broiler chickens from Sultanpur district were collected within 6 hours of death throughout the year (from May 2015 to April 2016). The birds were of different age group and of both sexes. After post mortem examination, all the gross lesions were recorded. For bacteriological study, materials from grossly suspected birds with *E. coli* infection were collected. The pieces of liver, lungs, heart, intestine, and spleen were aseptically collected from the dead birds showing gross pathological lesions of suspected *E. coli* infection i.e. yellow/white sero-fibrinous covering over liver, heart, and peritoneum, congestion of lung and heart, intestinal haemorrhages and necrotic foci on liver. They were kept in sterile vials separately and brought to the Veterinary Microbiology Laboratory under ice coverage for further study. The method described by Cruickshank *et al.*, (1975) was used for the isolation of *E. coli* and the isolates were identified on the basis of morphology, motility, and colony characteristics and bio-chemicals properties as per the method of Edwards and Ewing (1972).

## Results and Discussion

Samples were collected from the different farms of two districts of Eastern Uttar Pradesh. A total of 178 dead birds were collected from Faizabad district and 151 dead birds were collected from Sultanpur district, suspected to be suffering from colibacillosis. In the present study total 329 (178+151) naturally dead birds were brought to the departmental laboratory for details pathological observation and bacteriological isolation. Postmortem examination was conducted for all birds and different internal organ were examined carefully for presence of gross lesion.

The gross changes of liver were found as enlarged, congested and covered with thick yellow/white serofibrinous covering. The serofibrinous membrane covered the liver either partially or completely that differs with degree and severity of infection (Fig. 1 and 2). In intestine hemorrhages, congestion

and edematous swelling were noted. Heart was also covered with thick yellow/white serofibrinous covering (Fig. 1 and 2). In severe *E. coli* infection the serofibrinous membrane covered all the visceral organs continuously (Fig. 1 and 2). The gross lesions observed in the present study corroborated with the findings of Tonu *et al.*, (2011), Daud *et al.*, (2014) and Parwez *et al.*, (2015).

Among tested 178 and 151 samples from Faizabad and Sultanpur, 95 (53.37%) and 44 (29.13%) were found to be suggestive of *E. coli* giving typical large (2-3mm) lactose fermenting pink coloured colonies on Mac Conkey's agar plate (Fig. 3). They again produced characteristic 'metallic sheen' on EMB agar (Fig. 4). On Gram staining during morphological study, they produced pink coloured, rod shaped ones (that is gram negative) with characteristic arrangement. These finding were in accordance with Edward and Ewing (1972).

**Table.1** Result of bio-chemical reactions shown by *E. coli* isolates of Faizabad district

Bio-chemical reaction	Positive		Negative	
	Number	Percentage	Number	Percentage
Indole test at 37 <sup>0</sup> C	95	100	-	-
Methyl Red	95	100		
Vogas Proskauer	-	-	95	100
Citrate utilization	2	2.11	93	97.89
Urase activity	3	3.16	92	96.84
T.S.I. Test	95	100	-	-
H <sub>2</sub> S Production	-	-	95	100
Nitrate Reduction	95	100	-	-
Motility	91	95.78	4	4.22
D-glucose	95	100	-	-
Lactose	95	100	-	-
Sucrose	68	71.57	27	28.43
Mannitol	95	100	-	-
Dulcitol	71	74.73	24	25.27
Adonitol	2	2.10	93	97.9
Salicin	57	60	38	40

**Table.2** Result of bio-chemical reactions shown by *E. coli* isolates of Sultanpur district

Bio-chemical reaction	Positive		Negative	
	Number	Percentage	Number	Percentage
Indole test at 37 <sup>0</sup> C	<b>44</b>	<b>100</b>	-	-
Methyl Red	<b>44</b>	<b>100</b>	-	-
Voges Proskauer	-	-	<b>44</b>	<b>100</b>
Citrate utilization	-	-	<b>39</b>	<b>88.63</b>
Urease activity	-	-	<b>39</b>	<b>88.63</b>
T.S.I. Test	<b>44</b>	<b>100</b>	-	-
H <sub>2</sub> S Production	-	-	<b>44</b>	<b>100</b>
Nitrate Reduction	<b>44</b>	<b>100</b>	-	-
Motility	<b>44</b>	<b>100</b>	-	-
D-glucose	<b>44</b>	<b>100</b>	-	-
Lactose	<b>44</b>	<b>100</b>	-	-
Sucrose	<b>32</b>	<b>72.72</b>	<b>12</b>	<b>27.28</b>
Mannitol	<b>38</b>	<b>86.36</b>	<b>6</b>	<b>13.64</b>
Dulcitol	<b>38</b>	<b>86.36</b>	<b>6</b>	<b>13.64</b>
Adonitol	<b>11</b>	<b>25</b>	<b>33</b>	<b>75</b>
Salicin	<b>39</b>	<b>88.63</b>	<b>5</b>	<b>11.36</b>

**Fig.1** Colibacillosis affected bird showing thick white serofibrinous membrane that covers the visceral organs continuously



**Fig.2** Colibacillosis affected bird showing pericarditis, perihepatitis, and congestion of liver, heart, and intestine



**Fig.3** Showing characteristic pink colour colonies given by *E. coli* strain on Mac Conkey's agar plate



**Fig.4** Showing characteristic “metallic sheen” given by *E. coli* strain on Eosin Methylene Blue agar plate



The presumptive *E. coli* isolates had shown typical bio-chemical reactions. This confirmed these isolates. All these isolates (from district Faizabad) showed typical biochemical reaction which were positive to Indole test, M.R. test, T.S.I. agar test, nitrate reduction test and negative to VP test and H<sub>2</sub>S production test (Table 1). Among 95 *E. coli* isolates 97.89% were negative to citrate utilization test and 96.84% to urease activity test. Out of 95 isolates of *E. coli* 91 were motile. They all fermented D-glucose, lactose, manitol. Among 95 isolates of *E. coli*, 68 (71.57%) isolates, 71(74.73%) isolates and 57 (60.00%) isolates had shown fermentation of sucrose, dulcitol and salicin respectively. Only two isolates fermented adonitol. The isolates from the farms of Sultanpur also showed the typical reaction (Table 2). They were positive to Indole test, MR test, TSI agar test, nitrate reduction test. They had shown 100% motility and negative to VP and H<sub>2</sub>S production test. They had shown 88.63 % (39 birds) negative result to citrate utilization and urease activity test. They all fermented D-glucose and

lactose. Among the 44 isolates of *E. coli* 72.72% (32) isolates, 86.36% (38) isolates, 86.36% (38) isolates, 25% (11) isolates and 88.63% (39) isolates had shown fermentation of sucrose, mannitol, dulcitol, adonitol and salicin respectively.

In Faizabad district overall incidence of colibacillosis was 53.37% (that is 95 samples were positive out of 178 samples) which was similar to the reports of Phukan (1988) who reported overall 50.84% incidence of *E. coli* infection. In Sultanpur district, 29.13% birds (that is out of 151 samples, 44 samples were positive) for *E. coli* infection which was almost similar with the reports of Raji *et al.*, (2003) who reported 30-40% mortality due to colibacillosis in the broiler industry in Nigeria. The incidence in the present study was also similar with the findings of Chandra *et al.*, (2008) and Ashraf *et al.*, (2015) in the broiler chicken.

The present investigation throws light on the incidence and gross pathological changes due

to *E. coli* infection in broiler chicken. Many stress factor that lowered body defence mechanism, bad sanitary practice, poor ventilation, high density of chicken etc. can induce *E. coli* infection as *E. coli* is a common inhabitant of intestinal tract at a concentration of 10<sup>6</sup>/gm of faeces. So, this infection can be prevented by regular preventive treatment using antimicrobial agents along with improved sanitation, hygiene and better husbandry practice.

## References

- Ashraf, A., Abd El Tawab, A. A. Maarouf, Samir, A. Abd El Al, Fatma, I. El Hofy and Emad, E. A. El Mougny (2014). Detection of some virulence genes of avian pathogenic *E.coli* by polymerase chain reaction. *Benha Vet. Med. J. Vol.* 26 (2):159-176.
- Ashraf, A., Tawab, A. E., Samir, A. Abd El Aal., Ebstisam, M. mazid and Doaa, A.El. Morsey. (2015). Prevalence of *E. coli* in Broiler chickens in winter and summer seasons by application of PCR with its antibiogram pattern. *Benha Vet. Med. J.*, 29 (2): 119-128.
- Chandra B., Niyogi D., Ghosh C. K., Shukla V. K., Joshi R. K., Jana S. and Mukhopadhyay S. K. (2008): Incidence and pathomorphology of spontaneous *Escherichia coli* infection in broiler birds in two districts of West Bengal. *J. Interacad.*, 12(4): 521-524.
- Cruickshank, R., Dugid, J.P., Marmion, B.P. and Swain, R.H. (1975). *Medical Microbiology*. 12<sup>th</sup> edn. Vol. II. Churchill Livingstone, Edinburgh, London and New York.
- Daud, N.H.A., Hatin, N. N., Paan, F. H., Kyaw, T., Khaing, A. T., Abba, Y. and Abdullah, F. F. J. (2014). An outbreak of colibacillosis in a broiler farm. *J. of Ani. and Vet. Adv.* 13(8): 545-548.
- Edwards, P.R. and Ewing, W.H. (1972). *Identification of Enterobacteriaceae*. 3rd edn. Burgess Publishing Company, Minneapolis, Minnesota. 55: 415.
- Ewers, C., Janssen, T. and Wieler, L.H. (2003). Avian pathogenic *Escherichia coli* (APEC). *Berl-Munch. Tierarztl. Wochenschr.* 116 (9-10): 381-395.
- Kabir, S.M.L. (2010). Avian Colibacillosis and Salmonellosis: A closer look at epidemiology, pathogenesis, diagnosis, control and public health concerns. *Inter. J. Env. Res. Pub. Health.*, 7: 89-114.
- Parwez, S., Prakesh, A., Nimanapalli, R., Kumar, P. R., Kumar, M. and Rahman, S. (2015). A case report of colibacillosis in a broiler bird. *World. J. Phar. Res.*, 4(1): 854-856.
- Phukan, A. (1988). Studies on the etiology and treatment of colibacillosis in poultry. M. V. Sc. Thesis of Assam Agriculture University, Guwahati.
- Rahman, M.T., Saha, S. and Islam, M.A. (1999). Prevalence of salmonella in poultry feed of Mymensingh, Bangladesh. *The Bangladesh Vet.*, 16: 45-46.
- Raji, M, A., Adekeye, J. O., Kwaga, j. K. P. and Bale, J. O. O. (2003). *In vitro* and *in vivo* pathogenicity studies of *E. coli* isolated from poultry in Nigeria. *Israel J. Vet. Med.*, 58 (1): 21-28.
- Saleh, M., Seedorf, J, and Hartung, J. (2003). Total count of bacteria in the air of three different laying hen housing systems. *Dtsch. Tierarztl. Wochenschr.*, 110 (9): 94-97.
- Tonu, N. S., Sufian, M. A., Sarkar, S., Kamal, M.M., Rahman, M.H. and Hossain, M.M. (2011). Pathological study on colibacillosis in chickens and detection of *Escherichia coli* by PCR. *Bang. J. Vet. Med.* 9 (1): 17-25.