Multidrug Resistance Pattern of *E.coli* from Poultry

Thangavel Kandasamy[1], Tamilam Vembuvizhivendan[2] and Kannan[3]

[1]Assistant Professor and Head, [2]Assistant Professor,
Avian Disease Laboratory, Thalaivasal
[3]Assistant Professor, ILFC, Veterinary College and research Institute, Namakkal

Article Received on 20.05.2018 *Article Published on 10.07.2018*

Abstract

The emergence and diffusion of antibiotic-resistant bacteria has been a major public health problem for many years now. In this study, antibiotic resistance of Escherichia coli was investigated. The occurrence of colibacillosis was 65% in poultry. This study also revealed that the E. coli (78) isolates were more susceptible to levofloxacin (92.3%), enrofloxacin (88.4), ofloxacin (83.3%), gentamicin (73%), ceftriaxone (60.3) and neomycin (54%). Rationale use of these drugs may prevent the development of resistant isolates of E. coli in future. The E. coli isolates were resistant to amoxicillin (74.6%), cloxacillin (70.6%), tetracycline (60%) and amikacin (50%). Indiscriminate use, improper selection, improper dose, incorrect duration of antibiotics at farm level may be responsible for such a higher occurrence of resistance.

Keywords: *E.coli, antibiotic sensitivity, poultry, public health*

Introduction

Avian colibacillosis has been found to be major infectious diseases of all ages of layer and broiler birds. It occurs sporadically or enzootically in most of the countries of the world including India. The most important reservoir of *Escherichia coli* was the intestinal tract of animals, including poultry. In chickens, there were about 109 colony-forming units (CFU) of bacteria per gram of feces. Of these, 106 CFU were *Escherichia coli*, 10-15% of which were pathogenic serogroups. Coliforms transmitted between poultry and humans (Ojeniyi, 1989). Egg transmission of pathogenic *Escherichia coli* was common and responsible for high chicken mortality. Pathogenic coliforms were more frequent in the gut of the newly hatched chicks than in eggs from which they hatched (Harry and Hemsly, 1965). It causes severe economic losses of the poultry with morbidity and mortality varying in chicken from 10-50% or more (Vaillancourt *et al*., 1992) and decreased productivity in affected birds. Antimicrobial resistance, the ability of microorganisms (notably bacteria) to withstand antimicrobial agents (antibiotics), was an important and growing public health issue. However, over the years bacteria that were once controlled by these drugs have developed resistance so that common infections in humans can cause death in humanbeing. *Escherichia coli* was the primary causative agent of cellulitis, septicemia, and air sacculitis in poultry. *E. coli* was the most significant poultry bacterial pathogen. There were several antimicrobials that have been approved for treatment of *Escherichia coli* infections in poultry.

[1]Corresponding Author Email ID: ktvmicro@gmail.com

The long-term use of antimicrobials for therapy and growth promotion in animals selects for drug resistance in gram-negative pathogens (Ewers *et al*., 2009). Until today very little works have been performed in our country on antibiotic resistant pattern in poultry. Considering all these constraints, this pioneering work has been under taken to determine the present occurrence of multidrug resistance in colibacillosis in poultryr and find out the most effective antibiotics for treatment. The resistant pattern of *Escherichia coli* against antibiotics (Ceftriaxone, Ofloxacin, Amikacin, Gentamicin, Enrofloxacin, Levofloxacin, Amoxicillin, Oxytetracycline, Cloxacillin and Neomycin) was also observed in this study.

Material and methods

Samples collection

In this study, a total of 120 heart blood swabs were collected in sterile method (Ewing, 1986) randomly from diseased and freshly dead birds from different poultry farms located in and around Thalaivasal area of Salem district of Tamil Nadu. The predominant lesions revealed in postmortem examination were ascites, fibrinous pericarditis, splenitis, fibrinous perihepatitis, air sacculitis, and peritonitis. The collected heart blood swabs were subjected bacteriological analysis.

Bacteriological examination

Each heart blood swabs were directly inoculated in MacConkey broth and incubated for 18 h at 37°C. Then, a loopful from the previously inoculated broth was streaked onto MacConkey agar (Hi media) plates and incubated for 24 h at 37°C. Rose pink colonies were picked up and streaked onto Eosin Methylene Blue (Hi media) and incubated overnight at 37°C. The identification of *E. coli* isolates depends on the colony morphological characters, and biochemical tests results following Ewing (1986).

Antimicrobial sensitivity testing

It was performed by disc diffusion method (Bauer, 1966) using Muller-Hinton agar using antibiotic discs belongs to different antimicrobial classes including Ceftriaxone (30mcg), Ofloxacin (5mcg), Amikacin (30mcg), Gentamicin (10mcg), Enrofloxacin (10mcg), Levofloxacin (5mcg), Amoxicillin (30mcg), Tetracycline (30mcg), Cloxacillin (30mcg) and Neomycin (30mcg). Interpretation of the results was done following Clinical and Laboratory Standards Institute Guidelines (2011).

Results and discussion

Among 120 examined specimens, *E. coli* was identified and characterized in 65% (78/120) of the total examined samples based on phenotypical and biochemical characteristics (Ewing, 1986). Among the 120 chicken carcasses examined, expressing lesions of colibacillosis, bacteriological analysis of heart blood swabs got 78 positive cultures of *E. coli* (65%). In the remaining 34 cases (28.3%), bacterial cultures were negative for *E. coli*. The negative cultures may result from drug intervention before referring the cases to the laboratory as reported by Saberfar et al. (2008). The *E. coli* isolates were subjected to antibiogram

and for their antimicrobial susceptibility against 10 antimicrobial agents routinely used in the field was ascertained (Table-3).

Table 1: Antibiogram of *E. coli* isolates from poultry

S. No	Antimicrobial agent	No. *E. coli* isolates susceptible	susceptibility %
1	Amoxicillin	20	25.6
2	Cloxacillin	23	29.4
3	Amikacin	35	45
4	Ceftriaxone	47	60.3
5	Enrofloxacin	69	88.4
6	Ofloxacin	65	83.3
7	Levofloxacin	72	92.3
8	Gentamicin	57	73
9	Tetracycline	31	40
10	Neomycin	42	54

The antibiogram revealed that the *E. coli* isolates more susceptible to levofloxacin (92.3%), enrofloxacin (88.4), ofloxacin (83.3%), gentamicin (73%), ceftriaxone (60.3) and neomycin (54%). Rationale use of these drugs may prevent development of resistant isolates of *E. coli* in future. The *E. coli* isolates were resistant to amoxicillin (74.6%), cloxacillin (70.6%), tetracycline (60%) and amikacin (50%). These results were well accorded with Jhonson *et al.* (2007).

Table 2: Antibiotic sensitivity pattern of *E. coli*

S. No	Antibiotic sensitivity pattern	No. *E. coli* isolates
1	LE, EX, OF, GEN, CTR, AK, TE, N, AX, COX,	3
2	LE, EX, OF, GEN, CTR, AK, TE, N,	9
3	LE, EX, OF, GEN, CTR	22
4	GEN, CTR, AK, TE, N, AX, COX, LE	10
5	EX, OF, AK, N, AX, COX	2
6	EX, OF, GEN, N,	5
7	LE, OF, GEN, TE, N,EX	8
8	LE, EX, OF, CTR, AK,	3
9	TE, N, AX, COX, EX,LE, AK,	1
10	AK, N, AX, COX, EX, OF, LE	4
11	EX, OF, LE	9
12	LE, EX, AK, COX	3

Indiscriminate use, improper selection, improper dose, incorrect duration of antibiotics at farm level may be responsible for such a higher occurrence of multi drug resistance (Hassan *et al.*, 2013). Multi drug resistant *E. coli* isolates were found for 10 commonly used and market available antibiotics. The multi drug resistance may transfer to consumer via food and results in serious public health hazard as because Shivani *et al*., (2014) reported antimicrobial resistance is more frequent in pathogenic *E. coli* isolates. Hence in therapeutic decision these drugs should be used with caution and only after antibiotic sensitivity testing.

The risk of spreading antibiotic resistance from poultry to humans should be considered when there is contamination of animal products, especially chicken meat and eggs by bacterial strains resistant to most of antibiotics. Controlling such strains are effective with rational use of antibiotics.

Summary

The indiscriminate use of antibiotics in poultry without veterinary advice is s becoming increasingly common. This practice determines the selection of resistant bacteria and the increase in multidrug resistance. We observed the alarming rates for individual

and multiple antimicrobial resistance of *E. coli* against the majority of antibiotics routinely used in the field. This causes severe economic losses to the poultry industry, related to high mortality among birds.

References

Bauer, A. W., Kirby, W. M., Sheris, J. C. & Truck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. *Technical bulletin of the Registry of Medical Technologists* 36(3), 49-52.

CLSI. (2011) Performance Standards for Antimicrobial Susceptibility Testing, Twenty-First Informational Supplement. Vol. 31. Clinical and Laboratory Standards Institute M02-A10 and M07-A08, Wayne, PA.

Ewers, C., E. M. Antao, I. Diehl, H. C. Philipp, and L. H. Wieler. (2009). Intestine and environment of the chicken as reservoirs for extraintestinal pathogenic *Escherichia coli* strains with zoonotic potential. *Appl. Env. Microbiol.* 75:184–192.

Harry, E. G. & Hemsley, L. A. (1965). The relationship between environmental contamination with septicemia strains of *E. coli* and their occurrences in chickens. *The Veterinary Rec*ord 77, 241-245.

Hassan M and Alireza J (2013). Shiga toxin-producing Escherichia coli isolated from chicken meat in Iran: Serogroups, virulence factors, and antimicrobial resistance properties. Poultry Science 92: 1305-1313.

Johnson JR, Sannes MR, Croy C, Johnston B, Clabots C and Kuskowski MA (2007). Antimicrobial drug-resistant Escherichia coli from humans and poultry products, Minnesota and Wisconsin, 2002-2004. Emerging Infectious Disease 13: 838-846

Ojeniyi, A. A. (1989). Direct transmission of *Escherichia coli* from poultry to humans. *Epidemiology and Infection* 103, 513-522.

Saberfar E, Pourakbari B, Chabokdavan K, Taj DF (2008). Antimicrobial susceptibility of *Escherichia coti* isolated from Iranian broiler chicken flocks, 2005-2006. J. Appl. Poult. Res. 17:302-304.

Shivani C, Khurana SK and Mane BG (2014). Escherichia coli: Animal Foods and Public Health-Review. Journal of Microbiology, Immunology and Biotechnology 1: 31-46

Vaillancourt, J. P., Elfadil, A. & Bissailon, J. R. (1992). Cellulitis in poultry, *Canada Poultryman* 79, 34-37.