

ISOLATION, CHARACTERIZATION AND ANTIBIOGRAM OF PATHOGENIC *ESCHERICHIA COLI* RECOVERED FROM BROILER CHICKEN, RIYADH, SAUDI ARABIA

A. A. Al-Arfaj¹, A. M. Hessain^{2,4*}, H. A. Hemeg³, Kh. F. Mohamed⁴ and I. M. Moussa^{4,5}

^{2*}Department of Health Science, College of Applied Studies and Community Service, King Saud University, P. O. Box 22459 Riyadh11495, Saudi Arabia

¹Department of Botany and Microbiology, College of Science, King Saud University, Saudi Arabia.

³Department of Medical Technology/Microbiology, College of Applied Medical Science, Taibah University, Madinah, Saudi Arabia

⁴Department of Microbiology, Faculty of Veterinary Medicine, Cairo University, P. O. 2446, Cairo 14242 Giza, Egypt.

⁵Department of Botany and Microbiology, College of Science, King Saud University.

Corresponding Author E-mail address: ahessan@ksu.edu.sa ashgan319@yahoo.com,

ABSTRACT

Present study was conducted on 200 fecal samples of broilers chickens suffering from colisepticemia from poultry farms in Riyadh, Saudi Arabia. *Escherichia coli* strains were isolated from 88 (44%) samples. Out of *E. coli* strains, 79 (89.77%) belonged to O126: K71, O158: K-, O114: K90, O111: K58, O78: K80 and O119: K69. Whereas, O126: K71, O158: K- and O114: K90 were the most prevalent. The antibiotic sensitivity pattern showed that all isolates were totally resistant to ampicillin, chloramphenicol and streptomycin (100%). Resistance against erythromycin and nalidixic acid reached 81.01 % and was recorded in 20 strains O126: K71, 17 strains O158: K-, 15 strains O114: K90, 12 strains O111: K58, and decrease to 78.48% against ciprofloxacin in O126: K71, O114: K90, O111: K58, O78: K80 and O119: K69 strains. It should note that the recovered isolates were 84.81% intermediate to cephalixin and 53.16% sensitive gentamycin. This study highlights the high resistance of *E. coli* to antibiotics constitutes a threat to poultry industry in Saudi Arabia and the need for continuous surveillance of antibiotic sensitivity pattern of *E. coli* with a view to selecting appropriate therapy.

Key words: Antibioqram, avian pathogenic *Escherichia coli*, serotypes.

INTRODUCTION

Escherichia coli is a rod, Gram-negative, facultative anaerobic bacterium that colonizes the intestinal tract of humans (Drasar and Hill, 1974). *E. coli* is the causative agent to diverse diseases because of the different pathogenicity mechanisms and diseases. Among these diseases is colisepticaemia, which is characterized by bacteriaemia and organs colonization of *E. coli* (Barnes *et al.*, 2003).

For many years, antibiotic is randomly used for treatment purpose. This leads to indiscriminate use of antimicrobial drugs in poultry industry without prior testing that might have resulted antibiotic resistance causing a serious problem because it limits the therapeutic possibilities in the treatment of bacterial disease (Aarestrup, 2005).

Food from poultry products may transmit antimicrobial drug resistance of *E. coli* to humans. Acquired resistance to antimicrobial agents increasingly complicates the control of *E. coli* extra intestinal infections, which cause illness, mortalities and healthcare costs (Pitout *et al.*, 2005)

So the goal of our study is to identify the prevalent serotypes and their antibiogram. In the boiler chickens showed signs of colisepticemia collected from different farms located in Riyadh, Saudi Arabia.

MATERIALS AND METHODS

Samples: Two hundred fecal samples were collected under aseptic conditions from broilers chickens showed signs of colisepticemia from different poultry farms, located at Riyadh, Saudi Arabia at the period of 10th January to 10th June 2014. Samples were wrapped, kept in ice box and transferred to the laboratory immediately.

Isolation and identification of *E. coli*: Samples were primarily inoculated in pre-enrichment media then streaked on MacConkey agar medium and incubated aerobically at 37°C. After an overnight incubation, a part of single typical well isolated lactose fermenting colony was tested for sorbitol fermentation by culturing on sorbitol MacConkey agar and sorbitol phenol red agar media, then incubated at 37°C overnight. Morphological, cultural and biochemical examinations were carried out according to Murray *et al.* (2003)

Determining Serogroups: Serological identification was performed to the isolates of *E. coli* which were identified by biochemical tests using polyvalent and monovalent *E. coli* antisera (Welcome diagnostic antisera). Diagnostic *E. coli* O157 antisera (Difco) and H7anti- sera (Difco) were used for serological identification of *E. coli* O157: H7.

Antibiotic sensitivity test: Antimicrobial activity screening of isolated *E. coli* was performed was conducted using agar diffusion test (Bauer *et al.*, 1966). Mueller-Hinton (MH) agar (Difco) (25 ml) was soaped into Petri dishes. Sensitivity to antibiotic was studied with ampicillin, cephalixin, chloramphenicol, ciprofloxacin, erythromycin, gentamycin, streptomycin and nalidixic acid with concentration of 10, 30, 30, 5, 15, 10, 10 and 30 µg/disc, respectively. An amount of 0.5 freshly grown pure culture of *E. coli* was inoculated into the plates and allowed to spread gently over the entire surface with a glass rod spreader. After 1 to 2 minutes, the discs were placed at a distance of about 1 cm apart and incubated at 37°C for overnight. On the basis of the diameter of zones of inhibition produced around the antibiotic discs the inhibitory effect of the antibiotic to the growth of the culture was recorded according to NCCLS (2002).

RESULTS AND DISCUSSION

Antibiotic resistance and their effectiveness in treatment and controlling diseases pose a serious problem of concern. A total of 88 *E. coli* strains (44%) were isolated from 200 collected fecal samples. Barbour *et al.* (1999) reported 40.4% prevalence of *E. coli* in chicken which is closed to the present findings.

Among the 88 *E. coli* strains analyzed, a total of 79 (89.77%) of the isolates belonged to 6 different O serogroups and 9 isolate belonged to a non identified serogroup as follows, 20 strains (22.73%) belonged to O126: K71, 17 strains (19.32%) O158: K-, 15 strains (17.05%) O114: K90, 12 strains (13.64%) O111: K58, 8 strains (9.09%) O78: K80 and 7 strains (7.95%) O119: K69. From the obtained result, it is recognized that O126: K71, O158: K- and O114: K90 were the most prevalent.

In a study of Knöb *et al.* (2004), 11 serogroups were identified: O2, O6, O8, O21, O25, 46, O78, O88, O106, O111, and O143. Serogroup O6 was the most frequent, representing 62% of the total number of strains. Serogroups O2, O21, and O78, commonly found in poultry affected by colibacillosis, as well as Da Silveira *et al.* (2002) identified 2.3% of APEC serogroup O6 among isolates from chicks with omphalitis. While, Ewers *et al.* (2004), reported that 49.6% of the *E. coli* recovered from septicemia poultry in Germany could be grouped to serotypes O78, O2, and O1.

Table 1. Antimicrobial sensitivity test of *E. coli*.

Antimicrobial agents	O126:K71	O158: K-	O114: K90	O111: K58	O78: K80	O119: K69
Ampicillin	R*	R*	R*	R*	R*	R*
Cephalexin	I**	I**	I**	S***	I**	I**
Chloramphenicol	R*	R*	R*	R*	R*	R*
Ciprofloxacin	R*	I**	R**	R*	R*	R*
Erythromycin	R*	R*	R*	R*	I**	S***
Gentamycin	I**	I**	S***	S***	S***	S***
Streptomycin	R*	R*	R*	R*	R*	R*
Nalidixic acid	R	R	R	R	S	I

R* = Resistance. I** = intermediate. S*** = Sensitive

In 1999, chickens have been documented as the main source of antibiotic resistance in humans in Kingdom of Saudi Arabia (Al Ghamedi *et al.*, 1999) as well as in northern India in 1992 and Morocco in 1995. The in vitro sensitivity of 79 *E. coli* isolates was conducted against 8 antimicrobial drugs. The results are recorded in Table 1 and 2 revealed that all isolates were totally resistant to ampicillin. chloramphenicol and streptomycin (100%), Resistance against erythromycin and nalidixic acid reached 81.01 %. The resistance against erythromycin and nalidixic acid was recorded in 20 strains

O126: K71, 17 strains O158: K-, 15 strains O114: K90, 12 strains O111: K58, while the resistance decreased to 78.48% against ciprofloxacin in O126: K71, O114: K90, O111: K58, O78: K80 and O119: K69. Though, Prescott and Baggot (1993) reported good activity of erythromycin against some gram negative bacteria. Whereas Al-Ghamdi *et al.* (1999), found 34.7% resistant to ciprofloxacin. It should note that the recovered isolates were 84.81% intermediate to cephalixin and 53.16% sensitive gentamycin. Filali *et al.* (1985) reported that all the strains of *E. coli* were sensitive to gentamycin. Gyles

(2008) also reported the extreme resistant of Avian pathogenic *E. coli*, especially to sulfonamides tetracycline and streptomycin, and. The resistance to Furazolidone and Chloramphenicol was high, however, the fact that their use in veterinarian field is forbidden.

The major reasons for antimicrobial resistance are the misuse of antibiotic, poor sanitation as well as crowding which explain the high degree of resistance in *E. coli* in the present study (Vanden and Stobberingh, 1999).

Table 2. Antimicrobial sensitivity test and resistance pattern of *E coli* isolates.

Antimicrobial agents	Sensitive		Intermediate		Resistance	
	No.	%	No.	%	No.	%
Ampicillin	0	0	0	0	79	100
Cephalexin	12	15.19	67	84.81	0	0
Chloramphenicol	0	0	0	0	79	100
Ciprofloxacin	0	0	17	21.52	62	78.48
Erythromycin	7	8.86	8	10.13	64	81.01
Gentamycin	42	53.16	37	46.84	0	0
Streptomycin	0	0	0	0	79	100
Nalidixic acid	8	10.13	7	8.86	64	81.01

Conclusion: The information in this study highlights the ultimately needs for the development of the health policy of using antimicrobial agents in food of animal origin

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