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Prevalence of Bacterial Resistance to Commonly Used Antimicrobials among *Escherichia coli* Isolated from Chickens in Kerman Province of Iran

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The objective of this study was to determine antimicrobial resistance rate of *Escherichia coli* isolates from chickens against commonly used antimicrobials in veterinary medicine. A total of 200 faecal samples of chickens were analyzed for pathogenic *E. coli*. Isolates were subjected to antimicrobial resistance testing by the disc diffusion method. The microbial resistance of tylosin, oxytetracycline, linco-spectin, neomycin, sulfadiazine/trimethoprim, enrofloxacin, difloxacin, flumequine and forphenical were determined. Out of 200 bacterial isolates 96% were identified as *E. coli.* A high prevalence of resistance were observed in *E. coli* for commonly used antibiotics in poultry medicine, tylosin (100%), oxytetracycline (94.8%), sulfadiazine/trimethprim (80.7%), flumequine (79.7%), neomycin (79.2%), difloxacin (78.7%), enrofloxacin (65.5%). The least resistance rate of *E. coli* isolates were observed for florphenicol (4.2%) and linco-spectin (4.2%). Findings from this study indicates high resistance rate of *E. coli* to commonly used antibiotics, therefore, the use of antimicrobial agents should be restricted to treating infections and call for banning of antimicrobials as growth promoters in poultry industry.

Key words: *Escherichia coli*, microbial resistance, antibacterials

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INTRODUCTION

The use of antimicrobial agents in any venue, including therapeutically in human and veterinary medicine, or as prophylaxis for growth promotion in animal husbandry, ultimately exerts selective pressure favorable for the propagation of antimicrobial resistance bacteria[1,2].

Resistant bacteria from the intestines of food animals may be transferred to retail meat products resulting from faecal contamination during various stages of slaughter process (e.g., evisceration) and subsequent handling of animal tissues[3]. As a likely consequence, antimicrobial resistance phenotypes have been documented for zoonotic pathogens, including *E. coli*, *Salmonella* and *Campylobacter* species isolated from chicken, beef and pork[4]. *E. coli*, however, is an important cause of food borne illness[5]. In the United States, *E. coli* is estimated to cause 173,000 illness each year[5].

E. coli is the most common Enterobacterium and can serve as an indicator bacterium that easily acquires antimicrobial resistances commonly found in different animal species[5-7]. The use of *E. coli* as an indicator bacterium is also important, because changes in the resistance of this species may serve as an early warning system for resistance in potentially pathogenic bacteria[8]. Since the resistance rate of *E. coli* to commonly used antimicrobials were not determined in Iran, so this study was preformed to evaluate the profile of *E. coli* antimicrobial resistance in Kerman province of Iran.

MATERIALS AND METHODS

Two hundred faecal chicken samples which were referred to Kerman Poultry Diagnostic Laboratory during one year period (2003) were cultured for *E. coli* by routine laboratory methods, using blood agar and EMB agar (Eosin Methylene Blue) and incubated at 37°C for 24 h. One suspected *E. coli* colony from each sample (on the basis of colony size and morphology) was selected for identification and further studies. Antimicrobial susceptibility testing was carried out by agar disk diffusion method[9]. The antimicrobials tested were the followings:

Oxytetracycline, tylosin, linco-spectin, neomycin, sulfadiazine/trimethoprim (Sultrim), enrofloxacin, difloxacin, flumequine, and florphenicol. Susceptibility tests followed NCCLS break points[9]. *Escherichia coli* ATCC 25922 was used as quality control[9].

RESULTS AND DISCUSSION

A total of 192 *E. coli* isolates were obtained from 200 chicken faecal samples, which accounts for 96% of total isolates, followed by *Salmonella* (2.5%) and *Staphylococcus* (1.5%) strains (Table 1). The results of *in vitro* antimicrobial susceptibility testing of *E. coli* isolates to commonly used antibiotics in poultry medicine are shown in Table 2. High resistance rate were observed against tylosin (100%), oxytetracycline (94.8%), sulfadiazine/trimethoprim (80.7%), flumequine (79.7%), neomycin (79.2%), difloxacin (78.7%) and enrofloxacin (65.5%). The least resistant rate of *E. coli* isolates were observed for linco-spectin (4.2%) and florphenicol (4.2%).

The results of this study showed high resistance rate to the most commonly used antimicrobials which were used in poultry medicine such as tylosin, oxytetracycline, sulfadiazine/trimethoprim, flumequine, difloxacin and enrofloxacin. These findings are in agreement with data from several previous studies which have found that resistance to tetracycline derivatives, sulfa drugs and penicillins is common among *E. coli* isolates from food animals, meat and chickens[2,3,5,10-14].

All of these antimicrobials are commonly used and classically approved for veterinary use in Iran. Comparison of resistance rate with previous report of 1980 from Iran revealed that the resistance levels are still comparable or even higher than those of previous Iranian study[15]. Frequencies of 100% of tylosin resistance have been found in the *E. coli* isolates in our study. High resistance to tylosin have been reported for *E. coli* isolates from diarrhoeic calves in the Spain[16]. The resistance rate to tetracycline (94.8%) found to be comparable to previous levels in Iran (100%), Sweden (78-98%) and Spain (94-99%)[7,14,15] , but it was much higher than the resistance levels in Japan[6].

Table 1: The percentage of isolated bacteria from chicken samples

Bacterias	Isolated bacteria percentage
E. coli	96
Sal	2.5
Staph	1.5

E. Coli= Escherichia coli, Sal = Salmonella, Staph = Staphylococcus

Table 2: Resistance and sensitivity rate to 9 antimicrobials in 192 *E. coli* isolates from chicken samples

Drugs	Resistance (%)	Sensitivity (%)
Tylosin	100	0
Oxytetracycline	94.8	5.2
Sulfadiazine/Trimethoprim	80.7	19.3
Flumequine	79.7	20.3
Neomycin	79.2	20.8
Difloxacin	78.7	21.3
Enrofloxacin	65.5	34.5
Linco-spectin	4.2	95.8
Florphenicol	4.2	95.8

The high microbial resistance rate to sulfadiazine/trimethoprim (80.7%) was also comparable to resistance levels in other countries such as Saudi Arabia (57-99.1%), Sweden (62-90%), and Netherland (88-100%)[7,13,15].

The resistance rate to the most widely used fluoroquinolones such as flumequine (79.7%), difloxacin (78.7%) and enrofloxacin (65.5%) in this study was found to be much higher than the resistance rate to fluoroquinolones in the Spain (38%) , Japan (10%) and Morocco (100% sensitive)[5,7,18]. Neomycin resistance levels (79.2%) was found to be comparable to previous repots in Iran, but it was higher than those reported from Japan (32.6%), Spain (40%), Sweden (38-67%), and Morocco (100% sensitive to gentamicin)[5-7,18].

Resistance rate were found to be less than 5% for florphenicol (4.2%), and linco-spectin (4.2%), and these antimicrobials showed substantial activity against *E. coli* isolates from chicken samples. *E. coli* showed high sensitivity to florphenicol (99 -100%) in Spain which is in agreement with our results[16].The use of these antimicrobials is restricted to veterinary use in Iran, so such restricted use may have resulted in lower level of resistant against florphenicol and linco-spectin[6]. However, these results are in marked contrast with the results of the recent reports of some of other investigators which showed high resistance rate of *E. coli* isolates to lincomycin and spectinomycin in Japan, and Saudi Arabia[13,19,20]. The *E. coli* is commonly found in the intestinal tract of animals and can also be implicated in human and animal infectious diseases[2-4,6]. So the *E. coli* resistance may be transmitted to human if animal food are improperly cooked or otherwise handled[3,5,12,13]. The level of antibiotic resistance in *E. coli* represents as a useful indicator for resistance determination in bacterial populations and changes in the resistance of *E. coli* may serve as an early warning system for resistance in potentially pathogenic bacteria[5,8]. Since the results of this study showed the high resistance rate to commonly used antimicrobials in poultry industry, therefore, the use of antimicrobial agents should be restricted to treating infections and call for banning of antimicrobials as growth promoters in poultry industry[21,22].

Also the health authorities should provide strategies to improve rational drug prescription, through basic professional training, group discussion, supervision and continued medical education for the health professionals[23-25].

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