Antimicrobial Susceptibility of *Escherichia coli* Isolated from Iranian Broiler Chicken Flocks, 2005–2006

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Primary Audience: Researchers, Veterinarian Practitioners, Flock Supervisors, Microbiologists

SUMMARY

Escherichia coli is a major pathogen of worldwide importance in commercially produced poultry, contributing significantly to economic losses in chickens and turkeys. One hundred thirty-five cases in broilers were examined and cultured for isolation and antimicrobial sensitivity evaluation of *E. coli* between January 2005 and December 2006. In 103 cases (76.3%) *E. coli* were isolated and in 32 cases (23.7%) no *E. coli* growth was observed. Multiple resistances were seen in all isolates. All isolates were uniformly resistant to Tiamuline, Tylosin, and Bacitracin. We observed low levels of resistance to Gentamicin (12%), Kanamycin (0%), and Florfenicol (39%). Percentages of resistance to Tiamuline, Bacitracin, Tylosin, Colistin, and Erythromycin (≥99%); Tetracycline (96%); Oxytetracycline (93%); Flomequine (87%); Neomycine (87%); Lincospectin (79%); Difloxacin (78%); Enrofloxacin (76%); Cotrimoxazole (72%); Chloramphenicol (52%); and Ampicillin (49%) were determined. Our data show high levels of multiresistance among Iranian *E. coli* isolates. It seems that the pattern of antibiotic resistance of bacteria that are clinically important for the poultry industry should be monitored.

Key words: *Escherichia coli*, antimicrobial, susceptibility

2008 J. Appl. Poult. Res. 17:302–304
doi:10.3382/japr.2007-00102

DESCRIPTION OF PROBLEM

Escherichia coli is a major pathogen of worldwide importance in commercially produced poultry, contributing significantly to economic losses in chickens and turkeys. Colibacillosis begins, in general, with an infection of the upper respiratory tract, followed by septicemia. *Escherichia coli* is commonly found in the intestinal tracts of animals, but usually only certain pathogenic serotypes that show virulence factors (adhesive ability, aerobactin production, serum resistance, and presence of the ColV plasmid) cause disease conditions [1].

Antimicrobial therapy is an important tool in reducing the incidence and mortality associated with avian colibacillosis. However, resistance to existing antimicrobials is widespread and of concern to poultry veterinarians [1]. The fluoroquinolones are a new class of antimicrobials that

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exhibit excellent activity against gram-negative bacilli, but using these antimicrobials in poultry may cause cross-resistance with fluoroquinolones used for treatment of important human enteric infections [2]. The present study determines antimicrobial susceptibility among a collection of avian pathogenic *E. coli* recovered from diseased birds diagnosed with colibacillosis in Iran from January 2005 to December 2006.

MATERIALS AND METHODS

A total of 135 broiler chickens grown on different commercial farms from almost all parts of Iran, which were suspect colibacillosis cases, had been referred to the microbiology department of ViroMed Laboratory between January 2005 and December 2006. Bacteria were originally recovered from a variety of tissues including the air sacs, pericardial sac, heart, liver, trachea, joint cavity, bone marrow, blood, and spleen and plated on blood agar and MacConkey agar plates. All bacterial isolates were microbiologically identified by standard biochemical identification methods [3, 4]. Antimicrobial susceptibility testing was performed by the Kirby-Bauer disk diffusion method, which has been the predominant method used in Iran, in accordance with National Committee for Clinical Laboratory Standards guidelines at the time of the study [5, 6]. Quality control organisms were utilized routinely in the laboratory to ensure accurate performance of the susceptibility tests. For data analysis and presentation, bacterial isolates that showed intermediate susceptibility to an antimicrobial agent were categorized as resistant isolates. The antimicrobial agents selected for our analysis were antibiotics that were commonly included in the treatment of colibacillosis in Iran. Antimicrobial susceptibility results were rounded down if <0.5 and were presented as whole numbers if ≥0.5.

RESULTS AND DISCUSSION

Antibiotic usage is possibly the most important factor that promotes the emergence, selection, and dissemination of antibiotic-resistant microorganisms in veterinary medicine. In poultry flocks, inappropriate antibiotic therapy and using antibiotics as growth promoters may result in high antibiotic selection pressure. Therefore, poultry pathogenic bacteria contain a relatively high proportion of resistant isolates. These resistant bacteria cause problems in rearing poultry flocks and in human health. Hence, the Food and Drug Administration has emphasized that antibiotic-fed animals can produce and increase the spread of drug-resistant organisms to humans.

Table 1. Antimicrobial sensitivity of *Escherichia coli* isolates from Iranian broiler flocks

Antibiotics	Number of isolates	Percent of sensitivity
Gentamicin	99	88
Florfenicol	77	61
Tiamuline	98	0
Cotrimoxazole	100	28
Neomycine	95	13
Erythromycin	97	1
Flomequine	99	13
Lincospectin	96	21
Chloramphenicol	69	48
Tetracycline	92	4
Tylosin	98	0
Difloxacin	85	22
Ampicillin	99	51
Oxytetracycline	15	7
Colistin	5	0
Enrofloxacin	100	24
Bacitracin	8	0
Kanamycin	9	100

In this study from 135 colibacillosis cases in broiler chickens, *E. coli* were isolated from 103 cases (76.3%), and in 32 cases (23.7%) no *E. coli* were isolated. The negative cultures may result from drug intervention before referring the cases to the laboratory. The susceptibility pattern of each antimicrobial agent tested is shown in Table 1. The first group includes the antibiotics to which there were very high levels of resistance (70 to 100%). These are Tiamuline, Bacitracin, Tylosin, and Colistin (100%); Erythromycin (99%); Tetracycline (96%); Oxytetracycline (93%); Flomequine and Neomycine (87%); Lincospectin (79%); Difloxacin (78%); Enrofloxacin (76%); and Cotrimoxazole (72%). The second group includes the antibiotics to which there were moderate levels of resistance (30 to 70%). These are Chloramphenicol (52%), Ampicillin (49%), and Florfenicol (39%). The third group includes the antibiotics to which there were low levels of resistance (0 to 30%). These are Gentamicin (12%) and Kanamycin

(0%). All isolated *E. coli* showed resistance to 4 or more antibiotics, so multiple resistances was observed in all of our isolates. They are uniformly resistant to Tiamuline, Tylosin, Bacitracin, Colistin, and Erythromycin (≥99%).

As shown in Table 1, our isolates have good sensitivity to the Aminoglycosides, Kanamycin (100%), and Gentamicin (88%). Kanamycin and Florfenicol are relatively new antimicrobials in the Iranian poultry industry. Hence, it may take time to effectively select resistant bacteria. Because of some difficulties in using Gentamicin in poultry flocks, farmers are not interested to use it. Gentamicin is available in Iran for poultry flocks just for injection. Because injection of poultry flocks requires a specialized team, it would be expensive. Due to impaired quarantine in most Iranian poultry farms, these teams may introduce other diseases to the farm. Handling poultry flocks for injection may cause stress and consequently make the situation critical. Therefore, injection for broilers is not acceptable, especially in the time of colibacillosis, which is common in the late stage of rearing. So there is not any antibiotic selection pressure for Gentamicin. This may explains the high percentage of sensitivity against this antimicrobial agent.

In comparison with another study performed in northwest Iran in 2005, our data are almost the same [7]. Tabatabaei and Nasirian [8] reported 48, 56, and 44% resistance to Neomycine, Flomequine, and Enrofloxacin, respectively, in Iran in 2002. In comparison with our study, resistance to these antibiotics has increased more than 32%, but the rate of resistance to Chloramphenicol and tetracycline has not changed.

Comparing our data with studies performed in Europe and the United States show that Iranian isolates are relatively more resistant than European and US isolates [2, 9]. This can result from different reasons, including blind antimicrobial therapy, excessive usage of antimicrobials for prophylaxis, inappropriate treatment, and impaired quarantine systems in the Iranian poultry industry.

CONCLUSIONS AND APPLICATIONS

1. Our data indicate that antimicrobial resistance is an important factor challenging the poultry industry in our country. Unfortunately, there are no *E. coli* monitoring programs that control the trend of antibiotic resistance in Iran.
2. It seems necessary that an organization monitor the pattern of antibiotic resistance for any bacteria that is clinically important for poultry industry. Results of such monitoring can help us to choose an appropriate antimicrobial agent for therapeutic intervention.

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