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Emergence of Multidrug Resistant Strains of *Escherichia coli* Isolated from Urinary Tract Infections

***R Moniri[1], A Khorshidi[1], H Akbari [2]**

[1]*Dept. of Microbiology and Immunology, Kashan University of Mediacl Sciences and Health Services, Iran*
[2] *Dept. of Statistics, Kashan University of Mediacl Sciences and Health Services, Kashan, Iran*

Abstract

The emergence of multidrug resistant strains of *Escherichia coli* has complicated treatment decision and may lead to treatment failures. From April to November 2001 we prospectively evaluated the prevalence of resistance to trimethoprim-sulfamethoxazole (SXT), gentamicin, cephalothin, ciprofloxacin, and nitrofurantoin in 220 *Escherichia coli* isolates from patients with urinary tract infections in kashan, Iran. To assess the current breadth of multidrug resistance among urinary isolates of *E. coli.* of total isolates, 10.9% were resistant to three or more agents and considered multidrug resistant. Among the multidrug resistant isolates, 91.7% were resistant to SXT, 75% to gentamicin, 58.3% to cephalothin, 54.2% to ciprofloxacin and 45.8% to nitrofurantoin. The predominant phenotype among multidrug resistant isolates (29.2%) included resistance to SXT, gentamicin, and cephalothin. Rates of multidrug resistance were demonstrated to be higher among males (13.2%) than females (10.4%). There was no significant association between gender and reduced susceptibility. Continued local surveillance studies are urged to monitor emerging antimicrobial resistance and to guide interventions to minimize its occurrence.

Keywords: *Multidrug resistant, Escherichia coli, Urinary tract infections*

Introduction

Escherichia coli, the most common member of the family Enterobacteriaceae implicated in urinary tract infections, has showed increasingly resistant to antibiotics and caused some therapeutic problems (1, 2, 3). Trimethoprim-sulfamethoxazole (SXT) has been used widely as empirical therapy for urinary tract infections caused by *E. coli*. However, resistance to SXT among *E. coli* isolates from patients with urinary tract infections has increased, with a prevalence of resistance which is reported 30 to 50 percent (4). Resistance to SXT is generally associated with resistance to ampicillin, cephalothin, and tetracycline. Multidrug resistance may be transferred on a single plasmid (3, 5). Ciprofloxacin has been more active against *E. coli* strains than other commonly used agents such as SXT and ampicillin (6). Ciprofloxacin belongs to an important class of antibiotics for treatment of urinary tract infections. It is because of their value in human medicine that resistance to ciprofloxacin in *E. coli* has been viewed with great concern wherever it has begun to emerge in various countries (7). Only limited multidrug resistance among urinary tract isolates reported (5). Since resistance patterns among *E. coli* vary by region, local surveillance data needed to provide reliable information about emerging levels of antimicrobial resistance. The data generated should provide important information to assist clinician in making empirical therapeutic choices and to assist in the development of guidelines for drug use. MDR organisms can present substantial therapeutic challenges, and as such may pose greater public health problems than highly prevalent isolates that exhibit resistance to a single agent. The potential for commonly encountered gram-negative bacilli to acquire

***Corresponding author:** P. O. BOX: 87155-111, E-mail: moniri_re@yahoo.com

cross-resistance to several antimicrobial agents has been well documented (5, 9). For these reasons, and because multidrug resistance has not been commonly addressed in surveillance studies, we investigated the activities of the antimicrobial susceptibility status and also determined the frequencies of MDR phenotypes. The aim of the present study was to describe the current susceptibility patterns of *E. coli* to antibiotics used for urinary tract infections, as well as to descibe the prevalence of the MDR phenotypes of antibiotic resistance in Iran.

Materials and Methods

A total of 220 consecutive clinical isolates of *E.coli* obtained from community-acquired urinary tract infections, collected between April and Novomber 2001, were included in this prospective surveillance study. Strains were sent to central laboratory (Kashan University of Medical Sciences and Health Services, Kashan, Iran) to confirm identification of isolates by standardized methods. Identification of *E. coli* was based on Gram staining, indole, methyl red, vogesproskaur, citrate (IMViC), oxidase, and β-glucoronidase test results. Antimicrobial susceptibility tests were performed with all isolates by the disk diffusion method according to standards developed by the National Committee for Clinical Laboratory Standards (8) against antimicrobials commonly used therapy of urinary tract infections in Iran. The disks were used, manufactured by the Padtan-Teb Company, Iran. Their antibiotic concentration per disk were as follow: ciprofloxacin (5µg), gentamicin (10µg), cephalothin (30µg), and nitrofurantoin (300µg) and trimethoprim-sulfamethoxazole (1.25/23.75µg). Multidrug resistance strains (MDR) was defined as resistant to three or more of the antimicrobials tested. The statistical significance of the association between categorial variables was assessed by the chi-sqaure test. We used SPSS for windows (version 11) to perform analysis of clinical data.

Results

The overall rates of resistance for the 220 *E. coli* isolates analyzed are provided in Table 1. Of the agents tested, nitrofurantoin (7.3%) and cephalothin (10.9%) demonstrated the lowest rates of resistance, and SXT (51.8%) demonstrated the highest. The resistance rate to all five agents was high in this study. Among the 220 isolates that were tested against all five antimicrobials, 35.9% were susceptible to all the agents studied (Table 2) and 38.6% were resistant to a single agent, predominantly to SXT. MDR isolates accounted for 10.9% of the 220 isolates. The majority of MDR isolates (75%) were resistant to three antimicrobials, and these accounted for 8.2% of all isolates. Isolates were also identified that were resistant to four agents (20.8% of MDR isolates; 2.3% of all isolates) and all five agents (4.2% of MDR isolates; 0.5% of all isolates). All MDR phenotypes that were identified are listed in Table 3, with concurrent resistance to gentamicin, cephalothin, and SXT accounting for 29.2% of the MDR isolates. Resistance to SXT was a component 91.7% of the MDR isolates. Of nitrofurantoin and cephalothin resistant isolates, 68.8%, and 58.3% were MDR, in contrast to SXT, gentamicin and ciprofloxacin resistant isolates, of which 19.3 %, 41.9%, and 41.9% were MDR, respectively. Of the MDR phenotypes encountered, resistance to SXT, gentamicin, and cephalothin was the most prominent phenotypes among isolated *E. coli*. Antimicrobial resistance rates to individual agents and the percentage of isolates demonstrating an MDR phenotype were stratified by patient sex and summarized in Table 4. Gentamicin resistance rate was approximately twice as common among *E.*

coli isolates from males as those from females (P = 0.01). Although, the resistant rates for MDR *E. coli* do not show significantly differences between males and females, but trends toward higher rates of MDR *E. coli* among males (13.2 %) vs., females (10.4 %) were evident.

Table 1: Antimicrobial susceptibility results for 220 *E. coli* urinary tract isolates in Kashan-Iran in 2001

Antimicrobial agent	Susceptible Isolates (%-No.)	Resistant Isolates (%- No.)
Gentamicin	80.5 (177)	19.5 (43)
Cephalothin	89.1 (196)	10.9 (24)
Ciprofloxacin	85.9 (189)	14.1 (31)
Nitrofurantoin	92.7 (204)	7.3 (16)
SXT	48.2 (106)	51.8 (114)

Table 2: Resistance to one or more antimicrobial among 220 *E.coli* urinary tract isolate tested against all five antimicrobials in Kashan-Iran In 2001

No. of agents to which Isolates were resistant	Total % of Isolates (No.)	Gentamicin % (No.)	Cephalothin % (No.)	Nitrofurantoin % (No.)	SXT % (No.)	Ciprofloxacin % (No.)
0	35.9 (79)					
1	38.6 (85)	12.9 (11)	4.7 (4)		74.1(63)	8.2 (7)
2	14.5 (32)	43.8 (14)	18.8 (6)	15.6 (5)	90.6 (29)	34.4 (11)
3*	8.2 (18)	72.2 (13)	55.6 (10)	33.3 (6)	94.4 (17)	44.4 (8)
4*	2.3 (5)	80 (4)	60 (3)	80 (4)	80 (4)	80 (4)
5*	0.45 (1)	100 (1)	100 (1)	100 (1)	100 (1)	100 (1)

*10.9% (24 of 220)of isolates were resistant to three or more antimicrobials and defined as MDR.

Table 3: Antimicrobial resistance phenotypes of 22 MDR *E. coli* urinary tract isolates in Kashan-Iran in 2001

Antimicrobial resistance phenotype	No. of isolates	% MDR isolates	% Total isolates
SXT, gentamicin, cephalothin	7	29.2	3.2
SXT, ciprofloxacin, nitrofurantoin	3	12.5	1.4
SXT, ciprofloxacin, gentamicin	3	12.5	1.4
SXT, nitrofurantoin, gentamicin	2	8.3	0.9
SXT, ciprofloxacin, nitrofurantoin, gentamicin	2	8.3	0.9
Ciprofloxacin, , gentamicin, cephalothin	1	4.2	0.45
SXT, ciprofloxacin, cephalothin	1	4.2	0.45
SXT, nitrofurantoin, cephalothin	1	4.2	0.45
SXT, ciprofloxacin, gentamicin, cephalothin	1	4.2	0.45
SXT, ciprofloxacin, nitrofurantoin, cephalothin	1	4.2	0.45
Ciprofloxacin, nitrofurantoin, gentamicin, cephalothin	1	4.2	0.45
SXT, ciprofloxacin, gentamicin, cephalothin, nitrofurantoin	1	4.2	0.45

Table 4: Patient sex for 220 *E. coli* urinary tract isolates in Kashan-Iran in 2001

Patient Gender	Total % (No.)	Gentamicin % (No.)	Cephalothin % (No.)	Nitrofurantoin % (No.)	SXT % (No.)	Ciprofloxacin % (No.)	MDR % (No.)
Female	82.7(182)	15.9 (29)	11.5 (21)	6.6 (12)	51.6 (94)	13.2 (24)	10.4 (19)
Male	17.3(38)	36.8 (14)	7.9 (3)	10.5 (4)	52.6 (20)	18.4 (7)	13.2 (5)
Significant/ P.Value		P=0.01	N.S*	N.S*	N.S*	N.S*	N.S*

* Not significate

Discussion

The overall rates of resistance to SXT found in this study was significant and higher than those reported by others (5, 10, 11). For the past decades, trimethoprim-sulfamethoxazole or trimethoprim alone has been used widely as empirical therapy for urinary tract infections caused by *E. coli*. The emergence of resistance to SXT among *E. coli* urinary tract infections in Iran should come as no surprise, since it has been reported in southern Europe, Israel, and Bangladesh (12). The resistance rate to ciprofloxacin was higher than reported rates in previous studies (5, 10). The results of this study indicate that a ciprofloxacin-resistant phenotype without concurrent resistance to other antimicrobials was slightly higher than reported in previous studies (5). A decline in the activity of ciprofloxacin would be especially problematic in view of the ability of gram-negative bacilli to acquire resistance to all other classes of antimicrobials (13). The resistance rate to nitrofurantoin was higher than previous studies (5, 10, 14). A possible explanation for large proportion of *E. coli* isolated with reduced susceptibility to ciprofloxacin, SXT, and nitrofurantoin could be the misuse, and overuse of these drugs in humans, and the abundant use of these drugs in animals in Iran. The rate of resistance to cephalothin was lower than reported by other studies (5). The prevalence of resistant isolates was lower among females than males for all agents under the study except for cephalothin. The difference between *E. coli* strains isolated from males and females against gentamicin was statistically significant ($P = 0.01$).

The most striking finding from these data was the high prevalence of resistance to almost all antimicrobials in this study. It is unclear why *E. coli* from this area should manifest such high rates of resistance. Reasons may include differences in antimicrobial usage, infection control practices, and unrecognized factors. There is an urgent need to recognize all factors involved, control the use of antimicrobials in this area, and select the best strategies for the prevention and conrol the development of resistance.

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