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Risk Factor Analysis Among Trimethoprim-Sulfamethoxazole Resistant *Escherichia coli* Isolates

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Background: Trimethoprim-sulfamethoxazole (TMP-SMX) can be used for treatment of several infections including respiratory, renal, and gastrointestinal tract infections, skin and wound infections, septicaemia, and other infections caused by sensitive organisms.
Objectives: The aim of this study was to evaluate the risk factors for acquisition of TMP-SMX-resistant *Escherichia coli* strains among hospitalized patients in a teaching hospital of Sanandaj, Iran.
Materials and Methods: It was a case-control study on a patient, carrying a TMP-SMX-resistant *E. coli* strain. The control patient carried a TMP-SMX-sensitive *E. coli* strain. TMP-SMX resistance was determined using disk diffusion methods.
Results: Of 343 isolates, 197 (57.43%) were TMP-SMX-resistant. Using ventilator and catheter were risky for acquisition of TMP-SMX-resistant isolate (odds ratio (OR) = 3.037, 95% CI = 1.60-5.75, P < 0.000; OR = 2.93, 95% CI = 1.15-7.43, P < 0.013, respectively). There was significant correlation between days of staying in ward and TMP-SMX resistance (P < 0.003).
Conclusions: The main risk factors associated with TMP-SMX resistance were using of ventilator and catheter and days of staying in ward. There is need for more studies to evaluate the role of the factors to control the spread of drug resistance.

Keywords:Escherichia coli; Hospital; TMP-SMX; Risk Factor

1. Background

Antibiotic resistance is increasing among both hospitalized and outpatients because of excessive usage of antibiotics. One of the most prevalent bacterial families is *Enterobacteriaceae*, a group of aerobic Gram-negative bacilli including *Escherichia coli,* which are the most prevalent causes of urinary tract infections (1-3). Antimicrobial resistance among *Enterobacteriaceae* has been increasing in recent years. Many centers have reported resistant of *E. coli* isolates to many antibiotics such as ampicillin and trimethoprim-sulfamethoxazole (TMP-SMX), the first-line treatments for urinary tract infections (2). There are studies reporting the increasing resistancy of *E. coli* to TMP-SMX among urinary isolates (4). TMP-SMX is a prevalent antibiotic used for prophylaxis and treatment of infections. This widespread use is associated with increasing resistance rates to it. Most TMP-SMX resistance genes are in *Enterobacteriaceae* (5, 6). In 2011, WHO policy pointed on prevention of antimicrobial resistance (1). Drug-resistant isolates are big problems in the word health system and also in Iran. Determination of susceptibility of *E. coli* to antibiotic in laboratories has great importance in clinical outcomes and infection treatments. Several risk factors are associated with antibiotic resistance, including antimicrobial usage, previous hospitalization, severity of illness, surgery, and immunosuppression (7-9).

2. Objectives

The aim of the present study was to evaluate the risk factors involved in acquisition of TMP-SMX-resistant *E. coli* strains.

3. Materials and Methods

This study was conducted in Faculty of Medicine, Kurdistan University of Medical Sciences, Sanandaj, Iran. All samples were routinely cultured on MacConkey agar and blood agar plates. The blood samples were cultured in blood culture bottles. *E. coli* Isolates were identified at the species level, using standard biochemical tests and microbiological methods (10, 11). Only one isolate per patient was included in the study.

3.1. Microbiological Methods

TMP-SMX susceptibility tests were carried out by disk diffusion method on Mueller-Hinton agar plates (Merck, Germany). The results were expressed as susceptible or resistant according to the criteria recommended by Clinical and Laboratory Standards Institute (12).

3.2. Risk Factors Detection

To identify the risk factors associated with acquisition of

TMP-SMX-resistant strains, we performed a case-control study. Based on the study question, a case was defined as a patient carrying one TMP-SMX-resistant *E. coli* isolate. For each case patient, control was defined as a patient infected by a TMP-SMX-sensitive *E. coli* isolate during the same study period and with closest conditions as the cases. Data were collected by reviewing the patients' medical charts as well as the laboratory database. The following variables were collected and analyzed: age, gender, hospital location at the time of infection, underlying disease, clinical specimen, transfering from another hospital, trauma, admission to an intensive care unit, antibiotic exposure, duration of hospital staying and intensive care unit stay (in days) before infection, site of infection, species of the infecting organism and its antibiotic-susceptibility profile, and presence of a central venous catheter, urinary catheter, or mechanical ventilation. Finally, all the antimicrobial therapies were administered during 30 days, before the positive culture results were ascertained.

3.3. Statistical Analysis

We compared the characteristics of cases and controls by univariate analysis, and variables with P values of < 0.05 were entered in a logistic regression analysis. Chi-square or Fisher's exact test was used for comparison of dichotomous variables, and continuous variables were compared by the Wilcoxon test. Odds ratio (OR) and 95% confidence intervals (95% CI) were also calculated. All the tests were two-tailed and P values of < 0.05 were considered significant. The statistical analyses were performed using SPSS 16 for Windows.

4. Results

From a total of 343 isolates, we identified 197 (57.43%) TMP-SMX-resistant strains as cases and 146 (42.57%) TMP-SMX-sensitive strains as controls. The bacteria were isolated from outpatients (25.63%), ICU (19.86%), women's (17.69%), internal (15.16%), pediatrics (13.00%), and surgery ward (5.42%), respectively. TMP-SMX-resistant bacteria were most frequently isolated from urinary tract infections (63.96%). Other isolation sites included respiratory tract infections (18.27%), wound cultures (6.09%), and blood (5.58%) (Table 1).

To identify the risk factors for TMP-SMX resistance, we compared 197 case patients with TMP-SMX-resistant bacterial infections to 146 control patients with TMP-SMX-susceptible bacterial infections (Table 2).

Table 1. Distribution of Trimethoprim-sulfamethoxazole Susceptibility Pattern Based on the Sample Source [a,b]

Specimen	TMP-SXT		Total
	R	S	
Abscesses	0.51	0.00	0.29
Ascetic fluid	0.51	0.00	0.29
Blood	5.58	10.96	7.87
Catheter bleeding	0.51	0.00	0.29
CSF	1.02	0.00	0.58
Lung discharge	18.27	8.22	13.99
Other	1.02	0.68	0.87
Trachea	2.54	2.05	2.33
Urine	63.96	75.34	68.80
Vagina discharge	0.00	0.68	0.29
Wound	6.09	2.05	4.37
Total	100	100	100

[a] Abbreviations: CSF, cerebrospinal fluid; TMP-SXT, Ttrimethoprim-sulfamethoxazole; R, resistant; S, sensitive.
[b] Data are presented as %.

Table 2. Characterization of Risk Factors Associated with Occurrence of Trimethoprim-sulfamethoxazole Resistance Among *Escherichia coli* Strains[a]

	Study Group		P Value	OR (95% CI)
Variable	**Case, No. (%) (n = 197)**	**Control, No. (%) (n = 146)**		
Total [b]	85 (24.8)	46 (13.4)	0.018	1.65 (1.02-2.58)
Immunosuppressive treatment	6 (1.7)	1 (0.3)	0.128	4.55 (0.54-38.25)
Transfer to ICU	28 (9.9)	8 (2.9)	0.006	2.85 (1.26-6.47)
ICU hospitalization	34 (16.4)	10 (7.1)	0.003	2.83 (1.35-5.95)
Trauma	35 (10.2)	9 (2.6)	0.001	3.28 (1.52-7.08)
Ventilator use	48 (14.0)	14 (4.1)	0.001	3.03 (1.60-5.76)
Catheter use	22 (6.4)	6 (1.7)	0.013	2.93 (1.15-7.14)*
Blood transfusion	48 (14.0)	17 (5.0)	0.002	2.44 (1.34-4.46)
Nosocomial infection	44 (12.8)	13 (3.8)	0.001	2.94 (1.51-5.69)
Antibiotic use	10 (2.9)	4 (1.2)	0.212	1.89 (0.58-6.17)
Underlying disease	21 (6.1)	9 (2.6)	0.102	1.81 (0.80-4.09)

[a] Abbreviations: OR, odds ratio; CI, confidence interval; ICU, intensive care unit.
[b] All the patients were males.

5. Discussion

Antibiotic-resistant bacterial strains are big problems for physicians, as they usually rely on empiric treatments. Based on our search in database sites, this was the first analysis of risk factors for acquisition of TMP-SMX-resistant *E. coli* strains in our state. TMP-SMX is the oldest antimicrobial agent successfully used in treatment of a variety of infectious microorganisms. Resistance development to this antibacterial agent can cause clinical failure in treatment of infections and TMP-SMX-resistant pathogens are a major threat to infection treatment and control programs. We conducted a case-control study to determine the risk factors associated with acquisition of antibiotic-resistant strains. Based on epidemiological aspects, control patients were randomly selected from the same population source. History of ventilator usage was a risk factor for acquiring TMP-SMX resistance (OR = 3.037, 95% CI = 1.60-.75, P < 0.001). Resistance to most TMP-SMXs were located on integrons which can be transferred horizontally and play important roles in their dissemination (13). Therefore, genes can be transferred by ventilators and other devices in hospitals. However, exposure to immunosuppressive drugs was not significant in bivariate analyses (P < 0.128). In this study, another significant risk factor for acquiring TMP-SMX resistancy was use of catheter (OR = 2.93, 95% CI = 1.15-.14, P < 0.013). ICU hospitalization was a significant risk factor for acquisition of resistance strains (OR = 2.83, 95% CI = 1.35 to 5.95). There were no significant differences between case and control patients regarding demographic specification. No association was seen between having an underlying disease and isolation of resistant strain from patients in both groups. There was significant correlation between days of staying in ward and TMP-SMX resistance (P < 0.003).

In conclusion, these results suggested that ICU staying and ventilator usage were strongly related to acquisition of TMP-SMX-resistant strains. Emergence of resistance to broad-spectrum antibiotics among *E. coli* strains brings about adverse clinical outcomes; therefore, knowledge about the specific risk factors for resistance should aid in the selection of appropriate antibiotic therapies and designing appropriate control programs.

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