Academic Sciences Asian Journal of Pharmaceutical and Clinical Research

Vol 5, Suppl 2, 2012 ISSN - 0974-2441

Research Article

EXTENDED SPECTRUM B-LACTAMASES IN URINARY ISOLATES OF *ESCHERICHIA COLI* IN FIVE IRANIAN HOSPITALS

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Received:17 November 2011, Revised and Accepted: 28 January 2012

ABSTRACT

Background: Urinary tract infections caused by *Escherichia coli* have become a significant global public health problem with Kurdistan being no exception. Furthermore, the situation is worsening due to advent of increased antibiotic resistance.

Aims: The wide spread presence of antibiotic resistant *E. coli* in our environment necessitates regular monitoring of antibiotics susceptibility trends in the clinical isolates obtained from different regions to provide the basis for developing Local, National and International prescription programs that can be used for delineating guidelines to maintain the desired effectiveness of antibiotics.

Materials and Methods: Urine samples from five hospital microbiology laboratories were analyzed for isolation and identification of *E. coli*. *E. coli* PTCC 1533 was send to them as a positive control for antimicrobial susceptibility testing.

Antibiotic susceptibility of *E. coli* isolates were validated following the Kirby-Bauer disc diffusion technique using Muller Hinton agar. Screening test for ESBL was done according to the criteria recommended by CLSI.

Results: A total of 1257 *E. coli* strains were isolated from patients who suffer from UTI referred to five hospitals in Kurdistan province. The most resistant antibiotics tested against *E. coli* were penicillin, ampicillin, and amoxicillin. All the *E. coli* isolates were tested for ESBL production and 239 (19.02%) were found to be ESBL producers.

Conclusions - In conclusion, our study reinforces the necessity for appropriate use of antibacterial compounds, and with the technical ability we now have to see resistance at a genetic level, to monitor more detailed patterns of emergence.

Keywords: Antimicrobial resistance, E. coli, Urinary tract infection, Kurdistan

INTRODUCTION

Urinary tract infections (UTIs) having as etiologic agent *Escherichia coli* are common infections with an estimated annual global incidence of at least 250 million cases, being costly to both patients and health care funding system[1].

This important opportunistic pathogen that has shown an increasing antimicrobial resistance to most antibiotics[2-3] isolated from humans. It has been observed that antibiotic susceptibility of bacterial isolates is not constant, but dynamic and varies with time and environment[4].

The therapeutic steps during the treatment of UTIs, involves a short course of antimicrobial drug, such as antibiotics viz. ampicillin, chloramphenicol, colistin methane sulphonate, kanamycin, nalidixic acid, nitrofurantoin, streptomycin, norfloxacin, Trimethoprim-Sulfamethoxazole etc. The antibiotic resistance studies using urinary tract isolates of *E. coli* from patients have been reported to have shown increased resistance to certain antibiotics, particularly, Extended-Spectrum-Beta-Lactamase (ESBL). These organisms are now one of the main gram-negative species to cause infections with ESBL-positive bacteria in humans[5-6].

The increase of drug resistance among these organisms has made therapy of UTI difficult and has led to greater use of expensive broad spectrum antibiotics such as third generation of cephalosporin. Therefore, systematic monitoring of such resistance at local, national and international levels is recognized as an integral part of the control strategy by most national and international organizations including World Health Organization[7-8].

To satisfy the urgent need for an efficient surveillance system to monitor the possible impact of this policy, and to define current occurrence and phenotypes of multidrug-resistant (MDR) *E. coli* among UTI isolates, we launched a project to establish a province network for continuous monitoring of such resistance among *E. coli* isolated from urine at five hospital laboratories which are affiliated to Kurdistan University of Medical Sciences, Sanandaj, Iran.

MATERIALS AND METHODS

Five hospital microbiology laboratories were participated in the study. In charge of each hospital laboratory was asked to come for a meeting concerning the collection of urine specimen, isolation, identification and antimicrobial susceptibility procedure for *E. coli*, in order to have the same procedures in all the laboratories. *E. coli* PTCC 1533 was send to them as a positive control for antimicrobial susceptibility testing.

Isolation and identification of *E. coli*

Isolation and identification of *E. coli* to the species level was performed by standard methods[9].

Antibiotic susceptibility of *E. coli* isolates

Testing procedures were validated following the Kirby-Bauer disc diffusion technique using Muller Hinton agar[10]. For each isolate, antibiotic susceptibility was determined for 11 different antibiotics: Ampicillin, Amikacin, Tetracycline, Amoxycillin, Chloramphenicol, Co-trimoxazle, Nalidixic acid, Ciprofloxacin, Nitrofurantion, Cefotaxime, Imipenem, and Gentamycin.

Screening test for ESBL was done according to the criteria recommended by CLSI[10]. Briefly, we used CPM and CPM/clavulanic acid, CAZ and CAZ/clavulanic acid and CTX and CTX/clavulanic acid disks (MAST). After inoculating isolates in Muller- Hinton Agar (Merck, Germany) plates and 24 hours incubation, zones for compound disks larger than or equal to five mm compared with single disks, considered as producing ESBLs.

RESULTS

A total of 1257 *E. coli* strains were isolated from patients who suffer from UTI referred to five hospitals in Kurdistan province. The most resistant antibiotics tested against *E. coli* were tetracycline, amoxicillin, ampicillin and chloramphenicol (Table 1). All the *E. coli* isolates were tested for ESBL production and 239 (19.02%) were found to be ESBL producers (Fig. 1).

DISCUSSION

Antimicrobial resistance often leads to therapeutic failure of empirical therapy; therefore, knowledge of the local prevalence of pathogens and their antimicrobial sensitivity patterns is essential for clinicians in their routine work. Clinicians should also be aware of the sensitivity patterns in both neighboring and distant areas.

This study reveals the antibiotic susceptibility pattern and ESBL prevalence in *E. coli* isolated from patients suffering from UTIs in Kurdistan province, Iran. Majority (76.13%) of *E. coli* were isolated from female patients (Data not shown); an observation similar to other reports[11-12].

In the present investigation, high resistance of *E.coli* to numerous antibiotics was observed. These results are congruent to the results reported in Nigeria and Iran[13-14], who found more than 90% resistance of their *E. coli* isolates to ampicillin. The situation indicates a threat and a possibility that the *E. coli* could have become resistance to many more antibiotics to which it showed susceptibility earlier.

ESBL are now a significant problem in hospitalized patients throughout the world. The prevalence of ESBLs among clinical isolates varies greatly worldwide and patterns are rapidly changing over time[15].

By screening test for ESBL, 19.02% of *E. coli*, were found to be positive. Our study showed a lower percentage of ESBL producers when compared to reports of Babypadmini and Appalaraju[11], but was almost in concordance with Ava Behrooozi *et al* [14].

In conclusion, our study reinforces the necessity for appropriate use of antibiotics, and with the technical ability we now have to see resistance at a genetic level, to monitor more detailed patterns of emergence. Guidelines for the early phenotypic detection of ESBL in microbiology laboratories are needed.

Table 1: Antibiotic resistance pattern of *E. coli* strains isolated from urine specimens at five hospitals in Kurdistan Province.

Antibiotics	Resistance (%)
Ampicillin	84.97
Amikacin	42.33
Tetracycline	89.89
Amoxycillin	87.51
Chloramphenicol	86.0
Co-trimoxazle	75.02
Nalidixic acid	75.02
Ciprofloxacin	19.97
Nitrofurantion	19.97
Cefotaxime	32.54
Imipenem	10.03
Gentamycin	45.03
Carbanicillin	27.53

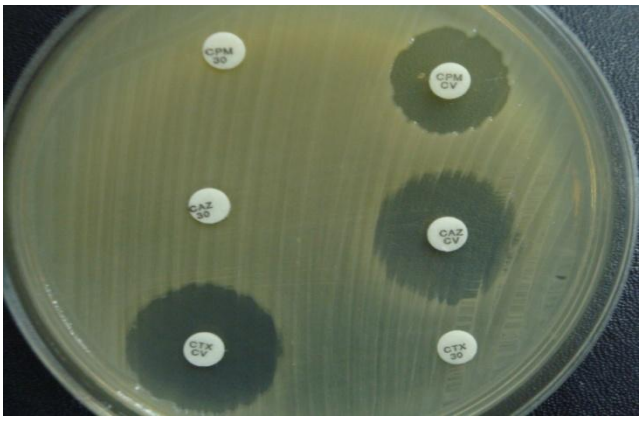

Fig 1: Phenotypic detection of ESBL by DDST among *E. coli strains* isolated from urine specimens at five hospitals in Kurdistan Province.

Acknowledgments

The authors wish to thank the following individuals for their assistance in the execution of this study: Dr. Estifaei F, Rashidi S and Amjadian P. This study was funded partly by a research grant from Kurdistan University of Medical Sciences, Sanandaj, Iran.

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