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Research Article

Antimicrobial sensitivity pattern among clinical isolates of *Escherichia coli* in tertiary care centre of Northern India

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ABSTRACT

Background: Rampant rise in resistance to various antimicrobials among clinical isolates of *Escherichia coli* is an issue of serious concern, especially in developing countries. It not only causes failure in treatment but also pose a health burden over the society at large.
Methods: Antimicrobial susceptibility testing was performed by Kirby-Bauer disc diffusion method on Muller Hinton agar as per Clinical and Laboratory Standard Institute recommendations over 100 isolates of E. coli. Isolate resistant to at least three classes of antimicrobial agents was considered as multidrug resistant. Isolate resistant to the three classes of antimicrobials along with carbapenem was considered extensively drug resistant.
Results: On performing the antimicrobial sensitivity pattern it was observed that *E. coli* isolates were maximally sensitive to ertapenem (56%), followed by imipenem (44%), meropenem (41%), piperacillin-tazobactam (23%), amoxicillin-clavulanic acid (21%), aztreonam (18%) among β-lactams group of antibiotics. 41% *E. coli* isolates were multidrug resistant and 11% were extensively drug resistant isolates.
Conclusions: Stringent measures should be undertaken to curb the spread of antibiotic resistance. Policies should be framed and implemented to stop over the counter sale of antibiotics.

Keywords: *E. coli*, Multidrug resistant, Urinary tract infection, Extensively drug resistant, Beta lactams, Antibiotic sensitivity

INTRODUCTION

Escherichia coli, one of the most significant members of the family *Enterobacteriaceae*, is most profuse facultative anaerobe of the human intestinal microflora.[1] It causes various intestinal and extra-intestinal diseases[2]and is one of the foremost cause of urinary tract and other opportunistic infections such as septicaemia, meningitis, peritonitis etc..[3,4] *E. coli* has acquired resistance to multiple antibiotics, therefore they pose a therapeutic challenge not only in the hospital settings, but also in the community.[5] The swift advent of antibiotic resistance among *E. coli* is a serious threat to the management of infectious diseases. β-lactam antibiotics are the most commonly used antimicrobials for empirical therapy. Production of β-lactamases is one of the mechanisms of resistance adopted by bacteria to develop resistance against β-lactams.[6] The various mechanisms of drug resistance in *E. coli* include efflux mechanism, porin deficiency along with production of various beta lactamases.[7-10]

Antimicrobial resistance has become a substantial public health concern particularly in third world countries.[11] Multidrug resistant pathogenic strains of *E. coli* are frequently seen in clinics signifying a major healthcare problem with increased morbidity and mortality.[12]

Keeping in view of this prospective, this study was conducted to know the antibiotic susceptibility in clinical

isolates of *E. coli* in this region and to know the prevalence of multidrug resistant isolates.

METHODS

The present study was conducted in Department of Microbiology, Pt. B. D. Sharma PGIMS Rohtak, Haryana, over 100 isolates of *E. coli* obtained from different clinical samples received in laboratory over a period of one year (2014-15). *E. coli* isolates were identified as per standard microbiological procedures. Antimicrobial susceptibility testing was performed by Kirby-Bauer disc diffusion method on Muller Hinton agar (MHA) as per Clinical and Laboratory Standard Institute (CLSI) recommendations. Antibiotic discs used in the study were procured from Hi-media Laboratories, Mumbai. American type culture collection (ATCC) strain viz *E. coli* ATCC 25922 strain was employed as control strain.[13,14] Discs of the following antimicrobial agents, were put up: ampicillin (10 µg), gentamicin (10 µg), amikacin (30 µg), amoxicillin-clavulinic acid (20 µg/10 µg), ampicillin-sulbactam (10 µg/10 µg), ticarcillin-clavulinic acid (75 µg/10 µg), cefuroxime (30 µg), cefepime (30 µg), ceftriaxone (30 µg), cefotaxime (30 µg), ciprofloxacin (5 µg), levofloxacin (5 µg), ertapenem (10 µg), imipenem (10 µg), meropenem (10µg), trimethoprim-sulfamethoxazole (1.25 µg/23.75 µg), aztreonam (30 µg), ceftazidime (30 µg), ofloxacin (5 µg), norfloxacin (10 µg) and nitrofurantoin (300 µg).

The diameter of the zones of complete inhibition (as judged by unaided eye) was measured to the nearest whole millimeter using a scale, which was held on the back of the inverted petridish. Any presence of small colonies (>1 colony) or a light film of growth within the zone of inhibition indicated resistance of the isolate to that antimicrobial agent.

The zones of inhibition were measured and were interpreted as sensitive (S), intermediate sensitive (IS) or resistant (R) according to the disc manufacturer information tables. An isolate was considered as multidrug resistant (MDR) if it was resistant to at least three classes of antimicrobial agents viz. all penicillins and cephalosporins (including inhibitor combinations), fluoroquinolones, and aminoglycosides. Isolate was considered extensively drug resistant (XDR) if it was resistant to the three classes of antimicrobials described above (MDR) and also resistant to carbapenems.[15]

The collected data was entered in Microsoft excel and then imported to SPSS (Statistical Package for Social Sciences Version 21, IBM Chicago USA) after data cleaning. The collected data was analyzed using appropriate statistical methods like percentages, proportions and chi square test. P value <0.05 was considered as statistically significant and <0.01 as statistically highly significant.

RESULTS

Sex wise distribution of patients with *E. coli* infection showed that males formed the majority (58%). Male to female ratio was found to be 1.38:1. The ages of patients with *E. coli* infection ranged from newborn to 88 years. Majority of patients from whom *E. coli* was isolated were in age group >60 years (20%), followed by 31-40 years (18%) and 41-50 years (16%).

Out of 100 clinical isolates of *E. coli*, majority of the isolates were from urine (47%) followed by pus (26%), blood (11%), stool (8%), sputum (5%) and body fluids viz. pleural, peritoneal, ascitic fluid etc. (3%). Fifty one (51%) isolates were from outdoor patients and 49 (49%) isolates were from indoor patients. Among the inpatients, majority of the isolates were from urine i.e. 36.7% (18/49), followed by pus 28.6% (14/49), blood 20.4% (10/49), body fluids 6.1% (3/49), stool and sputum 4.1% each (2/49). Among the outdoor patients, maximum isolates were from urine 56.9% (29/51), followed by pus 23.5% (12/51).

Table 1: Antimicrobial sensitivity pattern of 100 clinical isolates of *E. coli*.

Antimicrobial drug	*E.coli* isolates	
	No. of sensitive isolates	Percentage of sensitive isolates
β-lactams		
Ampicillin	6	6%
Ceftazidime	11	11%
Cefepime	10	10%
Ceftriaxone	13	13%
Cefoxitin	10	10%
Cefuroxime	6	6%
Cefotaxime	8	8%
Aztreonam	18	18%
Imipenem	44	44%
Meropenem	41	41%
Ertapenem	56	56%
Amoxicillin-clavulanic acid	21	21%
Ampicillin-sulbactam	13	13%
Ticarcillin-clavulanic acid	12	12%
Piperacillin-tazobactam	23	23%
Fluoroquinolones		
Ciprofloxacin	16	16%
Ofloxacin	16	28%
Norfloxacin	22	38%
Levofloxacin	7	7%
Aminoglycosides		
Amikacin	23	23%
Gentamicin	12	12%
Others		
Trimethoprim-sulfamethoxazole	15	15%
Nitrofurantoin	25	53%

On performing the antimicrobial sensitivity pattern it was observed that *E. coli* isolates were maximally sensitive to ertapenem (56%), followed by imipenem (44%), meropenem (41%), piperacillin-tazobactam (23%), amoxicillin-clavulanic acid (21%), aztreonam (18%) among β-lactams group of antibiotics (Table 1).

Forty one (41%) *E. coli* isolates were multidrug resistant and 11% were extensively drug resistant isolates. P value was found to be 0.00018 which was statistically highly significant. Out of 41 MDR isolates, maximum numbers of MDR strains were from urine (43.9%) followed by pus (41.5%), blood (8.3%), body fluids (5.6%) and sputum (2.8%) samples.

DISCUSSION

E. coli are the major cause of urinary tract and other opportunistic infections.[16] The present study showed maximum rate of isolation of *E. coli* was from urine (47%), followed by pus (26%).Similar observations were made by Chika et al who concluded in their study that maximum *E. coli* isolates were from urine (85%), followed by wound swabs (12.5%).[17]

The antibiotic resistance in isolates of *E. coli* has been amplified during the last decade.[16] Rise of antibacterial resistance is responsible for the empirical therapy failures. Various mechanisms exist which create resistance to antibiotics. Three elementary mechanisms of resistance to all antibiotics are known, i.e., target alteration, reduced drug concentration and inactivation of the drug. These mechanisms alone or in combination contribute to resistance against the various antibiotics.[16] Self medication, over the counter sale of antibiotics and injudicious use of broad spectrum antibitics by medical practitioners especially in developing countries, are some of the factors responsible for alarming rise of antimicrobial resitance in pathogens.

On performing the antimicrobial sensitivity pattern in 100 clinical isolates of *E. coli*, it was observed that among beta lactam antibiotics, susceptibility rates of *E. coli* isolates to carbapenems was high i.e. 56% isolates were sensitive to ertapenem, followed by imipenem (44%) and meropenem (41%). It was observed that ertapenem was the most effective antibiotic for *E.* coli isolates. Cefuroxime and ampicillin was found to be least sensitive drugs in our study with sensitivity rate of 6% each. Tanvir et al observed in their study that *E. coli* showed 100% sensitivity towards imipenem, 99.3% to meropenem, 96.8% to piperacillin-tazobactam, 77.2% to ceftazidime and 67.1% to aztreonam.[18] No resistance to imipenem was observed by Al-salamy, Messai et al, Enwuru et al, Bora et al and Mirzaee et al.[19-23]

Antimicrobial susceptibility rates of *E. coli* isolates to aminoglycosides i.e. amikacin and gentamicin was 23% and 12% respectively. Our results were somewhat similar to Ranjini et al who observed sensitivity of 16.7% to amikacin among *E. coli* isolates.[24]

The difference in antimicrobial susceptibility rates of *E. coli* isolates as compared to other studies may be due to geographical variations. The use of antimicrobials in empirical therapy and treatment varies from place to place, so the levels of resistance may vary.

The prevalence of multidrug resistant *E. coli* isolates i.e. isolates showing resistance to at least three classes of antimicrobial drugs i.e. penicillins and cephalosporins (including inhibitor combinations), fluoroquinolones and aminoglycosides was observed to be 41%. Tanvir et al observed in their study that 65.5% of isolates were MDR i.e. resistance to more than 8 antimicrobials (belonging to three or < 3 different classes of antimicrobials).[18] High percentage of MDR isolates of *E. coli* has been observed by Ibrahim et al[25] (92.2%), Ranjini et al (82.6%) and Bora et al (80%).[24-22]

Out of 41 MDR isolates, maximum numbers of MDR strains were from urine (43.9%) followed by pus (41.5%), blood (8.3%), fluid (5.6%) and sputum (2.8%) samples. Niranjan & Malini also observed that majority of *E. coli* isolates (76.5%) from urine samples were multidrug resistant.[26]

CONCLUSIONS

This study concluded that multidrug resistant *E. coli* isolates are on rise and susceptibility to various antibiotics even to carbapenems is decresing which is a matter of concern. Drug-resistance surveillance programmes should be run in hospitals to keep a check on antimicrobial resistance. Policies should be implemented to curb over the counter sale of antibiotics and stringent measures should be undertaken to limit the spread of antibiotic resistance.

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