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RESEARCH ARTICLE

ANTIMICROBIAL SENSITIVITY PATTERN OF *ESCHERICHIA COLI* ISOLATED FROM URINE SAMPLES OF UTI PATIENTS AND ISSUES RELATED TO THE RATIONAL SELECTION OF ANTIMICROBIALS

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ABSTRACT

Antimicrobial resistance is not only increasing the healthcare costs, but also the severity and death rates from certain infections that could have been avoided by prudent and rational use of the existing and newer antimicrobial agents. Prudent and rational use of antimicrobial is possible by forming local, national and global wide Antibiogram. The present study is undertaken to prepare local antibiogram and to discuss general issues related to antimicrobials use.

Total 170 urine samples were processed for culture sensitivity testing. Identification of bacteria was done by gram staining. Isolation of the organisms was done by inoculation of sample on agar medium. After 24 hours of incubation each organism was identified on the basis of morphology of colony in culture media and biochemical reactions. Antibiotic sensitivity Testing was carried out on Muller Hinton Medium by Disc Diffusion Method following Kirby Bauer method.Out of 170 samples were processed, 136 organisms were isolated from urine which includes *Escherichia coli* (76), *Klebsiella* (34), *Pseudomonas* (12), and *Staphylococci* (14). *E.coli* is most common organism (55.9 %). *E.coli* is most sensitive to piperacillin/tazobactam and gatifloxacin. An attempt has been made in this study to recognize the *E.coli* infection in patients of tertiary care hospital, Surendranagar, Gujarat; to record the antibiogram and probable drug of choice for *E.coli*. Considering the antibiotic susceptibility testing, cost, side effects and many other factors, gatifloxacin should be preferred for *E.coli* infection for UTI.

Keywords: UTI, *E.coli*, antibiotic susceptibility testing, antimicrobial resistance, rational selection of antimicrobials

INTRODUCTION

Antimicrobial agents are among the most commonly used and misused of all drugs. The inevitable consequence of the widespread use of antimicrobial agents has been the emergence of antibiotic resistant pathogens, fueling an ever-increasing need for new drugs. However, the pace of antimicrobial drug development has slowed dramatically, with only a handful of new agents, few of which are novel, been introduced into clinical practice each year. Reducing the inappropriate antibiotic use is thought to be the best way to control resistance [1].

The microbiology laboratory plays a central role in the decision to choose a particular antimicrobial agent over others. First, identification and isolation of the causative organism take place when the patients' specimens are sent to the microbiology laboratory. Once the microbial species causing the disease have been identified, a rational choice of the class of antibiotics

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likely to work in on the patient can be made [2].

Escherichia coli infection is an important cause of illness and death in infants in developing countries [3]. *E.coli* remained the most common causative agent of uncomplicated urinary tract infection (UTI) for many years with 75-90% causes of UTI infection[4-6].The other gram negative pathogens causing UTI are *Klebsiella* spp., *Proteus mirabilis* and *Pseudomonas aeruginosa*, however, *Enterococci* and coagulase negative *Staphylococci* are the most frequently encountered gram positive bacteria in UTI[7].To ensure appropriate therapy, current knowledge of the organisms that cause UTI and other infection, their antibiotic susceptibility testing is mandatory [8]. Due to rising antibiotic resistance among uropathogens, it is important to have local hospital based knowledge of the organisms causing UTI and their antibiotic sensitivity patterns [9].

Aims and objectives of this study are to find out the prevalence of *E.coli* infections in UTI in C.U. Shah Medical College and Hospital, Surendranagar, Gujarat, India and to determine the antimicrobial sensitivity pattern of *E.coli* isolated from urine sample of UTI patients as well as to find out rational antimicrobial agents.

MATERIALS AND METHODS

In the present study, 170 urine samples of urinary tract infection (UTI) patients were collected in Department of Microbiology from inpatient & outpatient department of C.U. Shah Medical College & Hospital surendranagar from period July 2008 to October 2008.

First step done was to isolate the organisms from these samples and then to study the culture susceptibility in common clinical isolates. Identification of bacteria was done by gram staining. Samples were inoculated on MacConkey, blood agar medium and Cystine Lactose Electrolyte Deficient (CLED) agar medium. After this, sample was inoculated on plates by four flame method. Inoculated culture plates were kept in the incubator at 37° C for 24 hours [10]. All the bacteria were identified using morphological, microscopy and biochemical tests following standard procedures described by Cowan and Steel and Cheesborough [11, 12].

Antibiotic sensitivity testing (AST) was done only for pathogenic bacteria. Antibiotic sensitivity was performed by Disc Diffusion Method of Bauer et al [13].The diameters of the zones of inhibition were measured by the unaided eye, including the diameter of the disc. AST of *Escherichia coli* (*E. coli)* towards different antibiotics was obtained. From this AST, antibiogram for *E. coli* was developed and on the basis of antibiotic sensitivity, cost effectiveness and ADR profile, appropriate antibiotic for treatment of *E. coli*, isolated from different urine samples was achieved.

RESULTS-

Total 170 urine samples were processed for culture sensitivity testing. Out of 170 samples, 136 organisms were isolated from urine which includes *Escherichia coli* (76), *Klebsiella* (34), *Pseudomonas* (12) and *Staphylococci* (14). *E.coli* is most common organism (76) i.e. 55.9 %.

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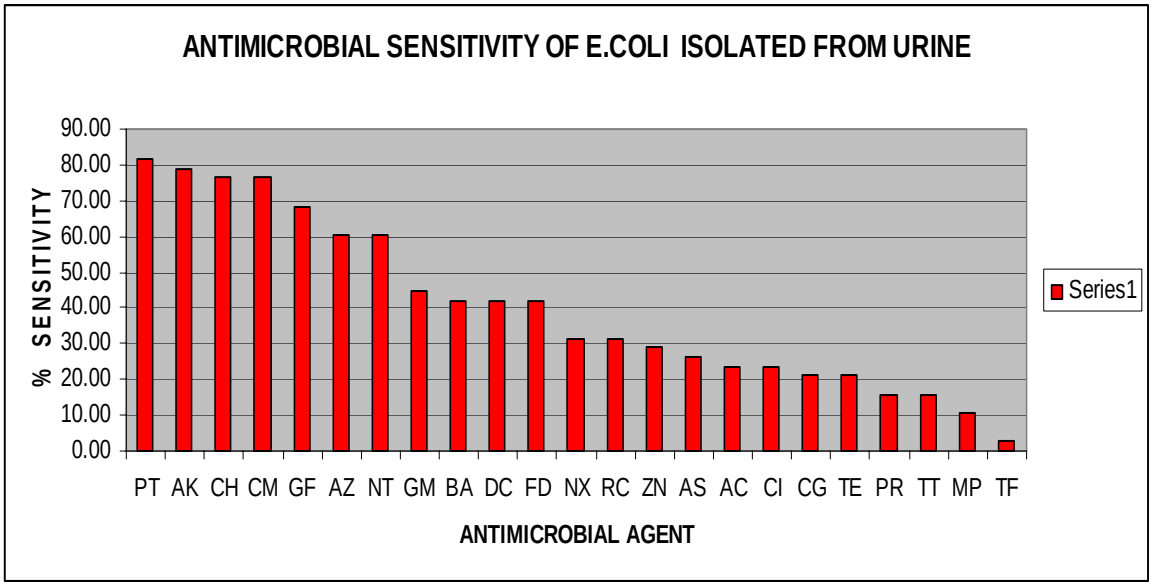
Figure 1 Antimicrobial sensitivity of *E.coli* isolated from urine samples of UTI patients -



abbr: - ac- aztreonam, ak- amikacin, as ampicillin/sulb az azithromycin, ba- Cotrimoxazole, cg- Cefipirome ch-Chloramphenicol, ci-Ceftizoxime), cm-Cefoperazone/Sulbactam, dc- Sparfloxacin, fd-nitrofurantoin, gf-gatifloxacin, gm-gentamicin, mp-meropenem, nt-netilmicin, nx- norfloxacin, pr-cephalexin, pt- piperacillin/tazobactam, rc-ciprofloxacin, te- tetracycline, tf- Teicoplanin, tt- Ticarcillin/clav, ZN- ofloxacin.

Table 1– Preferred drug of choice for *E.coli*, isolated from urine of the patient of UTI -

Sr no	Name of drug	% sensitivity.	Route	Price in Rs/10 tab/vial	Total duration of treatment	Total cost of treatment in Rs	ADR/ Toxicity of drug
1	Piperacillin/ Tazobactam	81	iv	238/ vial	QID × 7 days	6664	mild
2	Amikacin	79	iv/im	10/500 mg vial	15mg/kg in 3 divided doses for 5 days	90-100	Mild – mod.
3	Cefoperazone/ sulb	79	iv	52/ vial	1 gm inj. BD for 7 days	728	mild
4	Chloramphenicol	76.8	Oral/iv	40	500 mg QID×7-10 days	150-160	Mod-severe
5	Gatifloxacin	75	Oral/iv	50	500 mg OD ×7-10 days	35-50	mild

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Table-2 Comparison of antibiotic sensitivity of *E.coli* isolated from urine with other studies [14-16]

Workers	RC	NX	FD	NA	GM	AK	CN	AMP	COT	CF	TE
Uma M V et al-1987[14]	--	--	100 %	89%	78.90 %	--	--	--	--	--	--
Manish et[15] al-1995	61. %	78.8 %	80%	75.8 %	76.1%	78.3 %	--	--	--	90.8 %	--
Krishna et al[16]	70	57.5	24.4	44.9	73.1	54.1	57.7	73	74.4	71.8	73.1
Present study, 2008	31. 6	31.6	47.4	--	44.7%	79%	15.8 %	26.3	42.1	--	18.4

(abbr:-RC-ciprofloxacin, NX- norfloxacin, FD-nitrofurantoin, NA- nalidixic acid, GM-gentamicin, AK-amikacin, PR-cephalexin, AMP-ampicillin, BA-cotrimoxazole, CF-cefotaxime, TE-tetracycline).

Table-3 Comparison of antibiotic sensitivity of *E.coli* isolated from urine with previous study [17]

Serial Number	Name of antibiotics	% sensitivity previous study[17]	% sensitivity of antibiotic in present study
1	Amikacin	43.8	79
2	Cotrimoxazole	12	42
3	Gatifloxacin	16.7	68
4	Gentamicin	33.3	42
5	Norfloxacin	14.1	41

As shown in Figure 1, *E.coli* is most sensitive (81.6%) to piperacillin/ tazobactam. It is more than 60 % sensitive to amikacin and chloramphenicol, cefoperazone/sulbactam, gatifloxacin, azithromycin and netilmicin whereas is less than 50% sensitive for other antimicrobials.

Percentage antibiotic sensitivity of above 5 drugs is between 75-81 % (as shown in table-1), taking consideration of cost, adverse drug reaction and other factors, gatifloxacin should be preferred drug of choice. Alternatives can be amikacin, chloramphenicol and cefoperazone/sulb. Piperacillin/tazobactam should be reserved drugs for multidrug resistant organism.

Comparison of antibiotic sensitivity of *E.coli* isolated from urine of the patients of UTI of present study with study done by Uma M V et al[14], manish et al[15] and krishna[16] show that *E.coli* is less sensitive i.e. more resistant to ciprofloxacin, norfloxacin, cephalexin, ampicillin, cotrimoxazole, cefotaxime and tetracycline. It means resistance to these antimicrobials is increasing. Amikacin is more sensitive for

E.coli tetracycline in comparison to above studies (as shown in table-2).

Comparison of antibiotic sensitivity of *E.coli* isolated from urine of the patients of UTI of present study with study done by Swati Banerjee study [17] shows that *E.coli* is more sensitive to amikacin, cotrimoxazole, gatifloxacin, gentamicin and norfloxacin in present study (as shown in table-3).

DISCUSSION

In present study we observe that older drugs like ampicillin/sulbactam and chloramphenicol are becoming less resistant now but newer drugs like cephalosporins are getting resistant. It indicates that routine exposure of bacteria only to newly developed antibiotics eliminated resistance against older out of use antibiotics and present bacterial strains have grown sensitive to these outdated agents. Amikacin seems to be a promising therapy for *E.coli* and other Enterobacteriaceae family infection. The problem of increasing resistance to other Enterobacteriaceae family infection has limited the use of other classes of antibiotics like the fluoroquinolone, tetracyclines, macrolides and chloramphenicol.

overall resistance to various generations of cephalosporins was high on account of the production of extended spectrum β-lactamases (ESBLs) by the bacteria involved. Concurrent administration of a β-lactamase inhibitor such as clavulanate or sulbactam markedly expands the spectrum of activity of acid resistant penicillins like piperacillin and cephalosporins. The dose as well as the incidence of toxicity subsequently reduced if beta lactamase inhibitors are with piperacillin or cephalosporins.

In fact, the irrational and inappropriate use of antibiotics is responsible for the development of resistance of Enterobacteriaceae family infection to antibiotic monotherapy. Hence, there is a need to emphasize the rational use of antimicrobials and strictly adhere to the concept of "reserve drugs" to minimize the misuse of available antimicrobials. In addition, regular antimicrobial susceptibility surveillance is essential for area-wise monitoring of the resistance patterns. An effective national and state level antibiotic policy and draft guidelines should be introduced to preserve the effectiveness of antibiotics and for better patient management.

Hence, based on the observations of the present study, we recommend use of either piperacillin or cefoperazone along with β-lactamase inhibitors (clavulanate or sulbactam) against infection caused by Enterobacteriaceae family. Further, amikacin and netilmicin should be considered as a reserved drug for the treatment of severe nosocomial infections. Resistance to antimicrobial agents is an emerging problem that has required clinicians to alter empiric therapy for such diseases as bacterial meningitis and has prompted laboratory researchers to rethink testing strategies. The rate at which resistant organisms develop is related to their exposure to antimicrobial agents. Control strategies must include guidelines for prudent use of antimicrobial agents. The CDC has developed recommendations to help clinicians use these agents wisely to prevent the development and spread of resistant organisms [18, 19]. The emergence and spread of resistance can be reduced through appropriate or careful use of antimicrobial drugs and increasing awareness among the population to the hazards of inappropriate antimicrobial use through public health education campaign [20].

There is difference *in vitro* sensitivity of *E.coli* for antibiotic for different samples

which may be because of different strains of *E.coli* in different samples and different pharmacokinetics of different site of infection. Gatifloxacin, Chloramphenicol, cefoperazone plus sulbactam and piperacillin showed sensitivity of 85.7%.

In vitro sensitivity is an important factor yet other factors given below should also be seriously considered in selecting the antimicrobial agents for an infection. For example cost of drugs for complete treatment, route of administration (oral, parenteral etc.), sage (if the patient is neonate chloramphenicol is contraindicated) and pregnancy (tetracyclines are contraindicated). Other factors like allergic reactions to drugs like beta lactam antibiotic, kinetics of drugs and its concentration at the target site and mode and frequency of administration, bactericidal or bacteriostatic, efficacy/safety ratio, immunological status of the patient, ADR should also be considered.

CONCLUSION

Selection of drug of choice in any condition especially in infective diseases is not easy. We have to take into consideration the efficacy, safety, cost, pharmacokinetic, pharmacogenetics, convenience of administration and many other factors. In case of infectious diseases we have to pay attention to microbial sensitivity and resistance pattern to various antimicrobials. The sensitivity pattern cannot be the sole criteria. Because it is done *in vitro* and it fails to take into account the immunological status of the patient and clinical condition of the patient. An attempt has been made in this study to recognize the most common bacterial agent of infection in Surendranagar area and to record the antibiogram of the bacteria in this area. An attempt was again made to recognize the probable drug of choice based on antibiogram and some of the other factors namely the cost of treatment, mode of administration and adverse drug reactions. *E.coli* is most sensitive (82%) to piperacillin plus tazobactam if isolated from urine followed by gatifloxacin, amikacin and chloramphenicol.

Considering the antibiotic susceptibility testing, cost, convenience of administration, adverse drug reactions and many other factors, gatifloxacin should to be preferred followed by chloramphenicol and amikacin for *E.coli* isolated from the patients of UTI.

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