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Research Article

A STUDY ON ANTIBIOTIC SUSCEPTIBILITY AND RESISTANCE PROFILES OF BACTERIAL STRAINS ISOLATED FROM PATIENTS WITH URINARY TRACT INFECTION (UTI) AT KANCHIPURAM DISTRICT, TAMILNADU, INDIA

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ABSTRACT

Objective: Treatment of urinary tract infection (UTI) is becoming difficult due to the rising trend of antibiotics resistance. Current awareness on antibiotic resistance pattern is vital for appropriate treatment. A cross-sectional study to examine a current phenomenon of great concern among the medical communities in developing countries is raising Multiple Drug Resistant (MDR) organisms and the challenges of curing the infections. The aim of the study was the evaluation of antibiotic susceptibility and resistant patterns of UTI pathogens.

Methods: A total of 122 mid-stream urine samples were collected from urinary tract infected patients from age groups like less than 15 years, 15-40 years and above 40 years of both sex. Antibiotic sensitivity was determined using 23 chemical based antibiotics by Kirby-Bauer disk diffusion method.

Results: The organisms isolated from the patient's were *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus, Klebsiella pneumoniae*, *Enterococci, Enterobacter* and *Proteus vulgaris*. The isolated uropathogens showed resistant to Tetracycline (100%), Cefuroxime (94%), Penicillin (91%) and Ceftriaxone (87%) and sensitive to Imipenem, Chloramphenicol, Clindamycin (98%) and Vancomycin (95%).

Conclusion: Urinary pathogens exhibited resistance to regularly used antibiotics, hence the susceptibility and resistance patterns of urinary pathogens should be considered before starting treatment for UTI.

Keywords: Antimicrobial susceptibility, Bacteriuria, Multidrug resistance, Urinary tract infection, Uropathogenic bacterium.

INTRODUCTION

UTI is one of the most widespread bacterial infections in numerous developing countries where proper sanitation is not maintained effectively. It has been reported that up to 150 million people are infected annually worldwide [1]. UTIs detected in both hospitalized as well as outpatients. Hence it causes severe impact on the socioeconomic life of persons and in addition leads to a huge proportion of antibacterial drug consumption [2]. The symptoms of UTI are related to poor perineal sanitation, pregnancy, UT obstacle, urethral reflux, catheterization, sexual interaction and spermicidal contraception [3]. Among *E.coli* confirmed UTI patients, 69% were females and 31% were males. It showed that females are more liable to UTI than males and the commencement of the infection in female was high at the age of 21-30 years. The infection episode was elevated in March to October. The most resistance rates of *E.coli* from the urine culture were Amoxicillin, Doxycycline, Cephradine, Cephalexin, Cefixime, Cotrimoxazole, Ceftroxone and Ciprofloxacin, except Cefuraoxime because it was considerably sensitive [4]. Studies exposed that uropathogens create resistance against many antibiotics where *E.coli* is one among them. This is due to repeated misuse of antibiotics, insufficient dosage and easy accessibility of antimicrobial drugs [5, 6, 7, 8]. The antibiotic resistance cause difficulties to treat UTI and this fact requires proper use of antibiotics along with the formulation of new antibiotics [9]. The outcome of different assessment propose that the *E.coli* is accountable for the large percentage of UTI (53.24%), followed by other pathogens like *Enterococcus faecalis* (24.05%), *Proteus* sp.(19.537%), *Staphylococcus aureus* (19.206%), *Staphylococcus epidermidis* (7.8%), *Staphylococcus saprophyticus* (13.2%), *Klebsiella* sp. (11.96%),*Enterobacter* sp. (5.128%), *Pseudomonas aeruginosa* (3.4%), *citrobacter* sp. (1.92%) and *serratia marcescens* (0.8%). The most spreaded pathogen in female was *E.coli*, and for male it was *Proteus* sp. The proportion of UTI female was 81% and for male was 19%. The proportion of infection in married male was 10%, whereas in single unmarried was 9%, compared with a proportion of 55% in married female and 26% in unmarried female. For the treatment of UTI, the antibiotic Ciprofloxacin (5 µg) was found to be most efficient antimicrobial agent against all isolated bacterial pathogens, whereas Oxacillin (1 µg) was found to be less effective [10]. *Staphylococcus aureus* isolated from UTI were susceptible to Streptomycin, Ciprofloxacin and Cotrimoxazole, while *Escherichia coli* and Pseudomonas aeruginosa were resistant to Ampicillin and *P. aeruginosa* alone resistant to Ceporex [11]**.** UTI is generally cured with antibiotics, but this approach has some negative aspect. Due to the frequent use of antibiotic for UTI's and other infections, bacteria build up resistance to the antibiotics, making them less and no effective. Antibiotics can also affect the immunity, making more prone to many infections [12]. Patients were unconscious of the implications of drug resistance caused by maltreatment of antibiotics. They were also oblivious about antibiotic resistance and it causes. One third of the patients showed non observance towards their antibiotic treatment, hence they contribute to develop antibiotic resistance in the society. So edification about antibiotic resistance and significance of compliance to their antibiotic treatment should be addressed primarily in patients to control antibiotic resistance [13]. The tendency of rising resistance of UTI pathogens had been recorded in the latest years [14, 15, 16]. Therefore antibiotic resistance is a significant issue, hence the urine culture and antibiotic sensitivity assays should be carried out regularly in all UTI patients. Depending on the drug vulnerability, treatment should be planned with the apt antibacterial agent of less cost [4].

MATERIALS AND METHODS

Isolation and Identificationof pathogens

Midstream urine samples were preferred the isolation of the micro organisms. Hence it was inoculated into specific media and incubated at 37°C for one day. The bacterial colonies were identified morphologically and biochemically. The strains were two Gram-positive (*Staphylococcus aureus and Enterococci*) and five Gram-negative (*Escherichia coli, Pseudomonas aeruginosa, Klebsiella Pneumoniae, Enterobacter and Proteus vulgaris*) bacterium. In our study we have preferred MDR organisms responsible to cause of UTI. The organisms were sub-cultured, incubated at 37°C for 24 hours and stored at 4°C to maintain stock culture for further studies.

Chemical based antibiotic sensitivity assay by Kirby- Bauer Disc Diffusion method

Susceptibility and Resistance to 23 antimicrobial agents was analyzed by disc diffusion method according to the modified Kirby-Bauer technique. All the isolated strains were tested using following Antibiotics: Amikacin (AK) 30µg, Ampicillin (AMP) 10µg, Cefoperazone/Sulbactam (CFS) 30µg , Cefepime (CPM) 30µg, Ceftazidime (CAZ) 30µg, Ceftriaxone (CTR) 30µg, Ciprofloxacin (CIP) 5µg, Cefuroxime (CXM) 30µg, Cephalothin (CEP) 30µg, Chloramphenicol (C) 30µg, Clindamycin (CD) 2µg, Ciprofloxacin (CIP) 5µg, Erythromycin (E) 15µg, Gentamycin (GEN) 10µg, Imipenem (IPM) 10µg, Linezolid (LZ) 30µg, Ofloxacin (OF) 5µg, Oxacillin (OX) 1µg, Penicillin (P) 10µg, Piperacillin/Tazobactam (PIT) 100/10µg, Trimethopim/Sulfamethoxazole (COT) 1.25/23.75µg, Tobramycin (TOB) 10µg, Vancomycin (VA) 30µg and Tetracycline (TE) 30µg.

After incubation at 37°C for 24 hours the inhibition zone (in diameters) were measured. Isolates were considered multidrug resistant if they showed resistance to 3 or more of the tested antibiotics. The Multiple Antibiotic Resistance indices (MARI) were calculated as follows: MAR index for isolates = [Number of antibiotics to which the isolate is resistance / Number of antibiotics tested]. While MAR index for an antibiotics = [Number of antibiotics resistance to the isolates / (Number of antibiotics x Number of isolates)].

RESULTS

The multiple antibiotic resistant indices (MARI) calculated for *E. coli* was 0.92, 0.33 in one each strain, 0.67 in 8 strains, 0.75 in 5 strains, 0.42, 0.83, 0.58 in 3 strains and 0.17, 0.08 in 2 strains. The MARI for *Pseudomonas aeruginosa* were 0.20, 0.5 in one each strain, 0.70, 0.00 in 4 strains, 0.80, 0.60 in 3 strains and 0.10 in 2 strains. The MARI for *Staphylococcus aureus* were 0.20 in 16 strains, 0.10 in 12 strains, 0.30 in 9 strains, 0.50 in 6 strains, 0.40 in 4 strains and 0.00, 0.60 in 3 strains. The MARI for *Enterococci* were 0.25, 0.88 in one each strain and 0.38 in 2 strains. The MARI for *Klebsiella pneumonia* were 0.08, 0.33, 0.42, 0.50 in one each strain and 0.17, 0.67 in 4 strains. The MARI for *Proteus vulgaris* was 0.58, 0.17 and 0.08 in one each strain. The MARI for *Enterobacter* was 0.50 in one strain and 0.58 in 3 strains (Table 1).

Most of the isolated uropathogens were resistances to commonly used antibiotics such as Tetracycline (100%, MARI 0.000), Cefuroxime (94%, MARI 0.041), Penicillin (91%, MARI 0.040, Ceftriaxone (87%, MARI 0.038), Ampicillin (60%, MARI 0.027), Cefoperazone / Sulbactam (60%, MARI 0.026), Ofloxacin (57%, MARI 0.025), Cefepime and Ciprofloxacin (52%, MARI 0.023), Ceftazidime (51% MARI 0.022), Tobramycin (50% MARI 0.022), Gentamycin (42%, MARI 0.018),Trimethopim/Sulfamethoxazole (36%, MARI 0.016), Oxacillin (28%, MARI 0.012), Erythromycin and Piperacillin/Tazobactam (23%, MARI 0.010), Amikacin (11%, MARI 0.005), Cephalothin (8%, MARI 0.003), Vancomycin (5%, MARI 0.002), Imipenem, Chloramphenicol, Clindamycin and Linezolid (2%, MARI 0.001) (Table 2). Total of 23 antibiotics response of uropathogens were showed on figure 1.

Table 1: It shows MAR indexing of isolates against antibiotics

E1 to E40	*E. coli*
S1 to S53	*Staphylococcus aureus*
K1 to K12	*Klebsiella pneumonia*
Eb1 to Eb4	*Enterobacter*
Pa1 to Pa18	*Pseudomonas aeruginosa*
Ec1 to Ec4	*Enterococci*
Pv1 to Pv3	*Proteus vulgaris*

ISOLATES	R	S	MARI	ISOLATES	R	S	MARI	ISOLATES	R	S	MARI
E1	9	3	0.75	Pa14	6	4	0.60	S37	1	9	0.10
E2	5	7	0.42	Pa15	6	4	0.60	S38	5	5	0.50
E3	9	3	0.75	Pa16	7	3	0.70	S39	2	8	0.20
E4	2	10	0.17	Pa17	7	3	0.70	S40	1	9	0.10
E5	5	7	0.42	Pa18	1	9	0.10	S41	1	9	0.10
E6	2	10	0.17	S1	2	8	0.20	S42	3	7	0.30
E7	10	2	0.83	S2	0	10	0.00	S43	1	9	0.10
E8	11	1	0.92	S3	1	9	0.10	S44	1	9	0.10
E9	7	5	0.58	S4	6	4	0.60	S45	2	8	0.20
E10	7	5	0.58	S5	2	8	0.20	S46	2	8	0.20
E11	8	4	0.67	S6	3	7	0.30	S47	1	9	0.10
E12	10	2	0.83	S7	1	9	0.10	S48	0	10	0.00
E13	9	3	0.75	S8	3	7	0.30	S49	5	5	0.50
E14	8	4	0.67	S9	1	9	0.10	S50	3	7	0.30
E15	7	5	0.58	S10	2	8	0.20	S51	2	8	0.20
E16	8	4	0.67	S11	2	8	0.20	S52	2	8	0.20
E17	10	2	0.83	S12	4	6	0.40	S53	6	4	0.60
E18	8	4	0.67	S13	4	6	0.40	Ec1	7	1	0.88
E19	8	4	0.67	S14	1	9	0.10	Ec2	3	5	0.38
E20	8	4	0.67	S15	4	6	0.40	Ec3	3	5	0.38
E21	9	3	0.75	S16	5	5	0.50	Ec4	2	6	0.25
E22	4	8	0.33	S17	2	8	0.20	K1	2	10	0.17
E23	1	11	0.08	S18	1	9	0.10	K2	2	10	0.17
E24	9	3	0.75	S19	2	8	0.20	K3	6	6	0.50
E25	1	11	0.08	S20	2	8	0.20	K4	4	8	0.33
E26	5	7	0.42	S21	3	7	0.30	K5	2	10	0.17
E27	8	4	0.67	S22	2	8	0.20	K6	8	4	0.67
E28	8	4	0.67	S23	2	8	0.20	K7	5	7	0.42
Pa1	7	3	0.70	S24	5	5	0.50	K8	2	10	0.17
Pa2	1	9	0.10	S25	3	7	0.30	K9	8	4	0.67
Pa3	0	10	0.00	S26	3	7	0.30	K10	1	11	0.08
Pa4	0	10	0.00	S27	5	5	0.50	K11	8	4	0.67
Pa5	8	2	0.80	S28	3	7	0.30	K12	8	4	0.67
Pa6	8	2	0.80	S29	0	10	0.00	Pv1	7	5	0.58
Pa7	8	2	0.80	S30	4	6	0.40	Pv2	2	10	0.17
Pa8	2	8	0.20	S31	6	4	0.60	Pv3	1	11	0.08
Pa9	7	3	0.70	S32	2	8	0.20	Eb1	6	6	0.50
Pa10	0	10	0.00	S33	1	9	0.10	Eb2	7	5	0.58
Pa11	6	4	0.60	S34	5	5	0.50	Eb3	7	5	0.58
Pa12	5	5	0.50	S35	2	8	0.20	Eb4	7	5	0.58
Pa13	0	10	0.00	S36	3	7	0.30				

R = Resistance;S = Sensitive

Table 2: It shows MAR index of antibiotics against isolated uropathogens.

Antibiotics	R	S	MARI
Amikacin	7	58	0.005
Ampicillin	32	19	0.027
Cefepime	34	31	0.023
Cefoperazone/Sulbactam	39	26	0.026
Ceftazidime	27	26	0.022
Ceftriaxone	41	6	0.038
Cefuroxime	44	3	0.041
Ciprofloxacin	36	33	0.023
Gentamycin	29	40	0.018
Imipenem	1	64	0.001
Ofloxacin	70	52	0.025
Piperacillin/Tazobactam	15	50	0.010
Tobramycin	9	9	0.022
Cephalothin	4	49	0.003
Chloramphenicol	1	52	0.001
Clindamycin	1	52	0.001
Erythromycin	13	44	0.010
Linezolid	1	52	0.001
Oxacillin	15	38	0.012
Penicillin	52	5	0.040
Trimethopim/Sulfamethoxazole	19	34	0.016
Vancomycin	3	54	0.002
Tetracycline	4	0	0.043

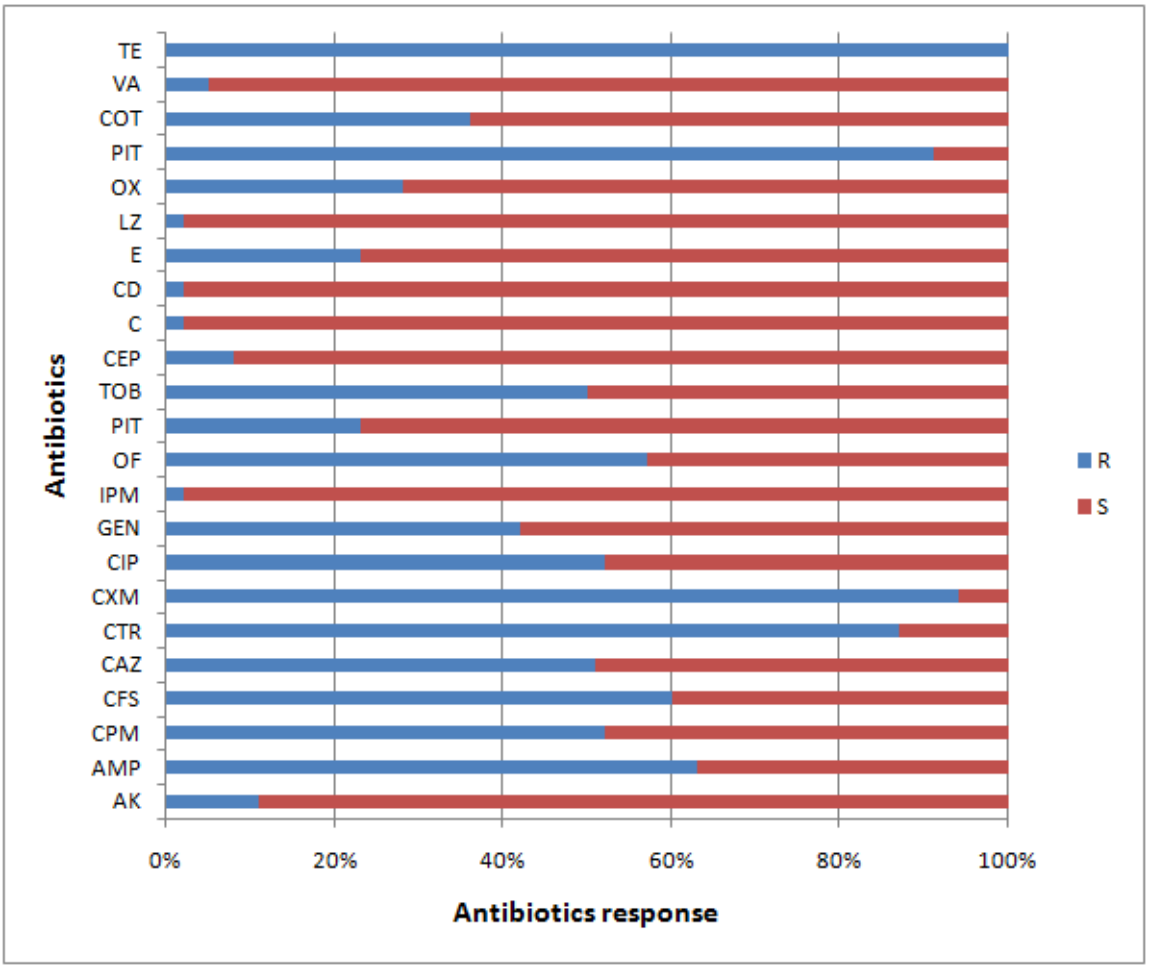

Fig. 1: It shows antibiotics response of uropathogens.

DISCUSSION

The maximum value of MARI for *E.coli*, *Pseudomonas aeruginosa, Staphylococcus aureus, Enterococci* and *Klebsiella pneumonia* were 0.92, 0.80, 0.60, 0.88 and 0.67 respectively.

The MARI for *Proteus vulgaris* and *Enterobacter* was 0.58. Some antibiotics showed maximum MARI against isolated uropathogens, for example Tetracycline, Cefuroxime, Penicillin, Ampicillin, Cefoperazone/Sulbactam and Ofloxacin were 100%, 94%, 91%, 63%, 60% and 57% respectively.

CONCLUSION

In our current study the MARI of 117 pathogens isolated from UTI were exposed to the antibiotics showing MARI except 5. Most of the uropathogens showed multiple antibiotics resistance, due to huge portion of the bacterial isolates being exposed to many antibiotics in the past. Our result shows information about the trend of increased antibiotics resistance of UTI pathogens at kanchipuram district, Tamilnadu, India, which may be due to geographic variation or indiscriminate or sublethal use of antibiotics. This information may useful for the

physician to prescribe most sensitive antibiotic to the patient for the appropriate treatment of UTI, also helpful to put off the random use of antibiotics and stop further expansion of bacterial drug resistance.

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