PREVALENCE OF UROPATHOGENS IN REPRODUCTIVE AGE GROUP FEMALES AND THEIR ANTIBIOTIC RESISTANCE PATTERN

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Abstract

Introduction: Urinary Tract Infection (UTI) remains the most common bacterial infection in human population. The prevalence of UTI is much higher in females as compared to males. The present study was planned with the aim to find out prevalence of uropathogens and their antibiotic resistance pattern in reproductive age group females.

Materials and Methods: The present study was carried out in department of microbiology, Jayoti Vidyapeeh Women's Universiy, Jaipur from Jan. 2020 to September 2020. Total 105 urine sample were processed for culture and sensitivity testing. Isolation of organism were done by inoculation of samples on Mac-Conkey Agar and Blood Agra media. After 24 hour of incubation at 37^0 C, each isolates were identified on the basis of morphology of colony in culture media, Gram stain, motility and biochemical reactions. Antibiotic sensitivity testing were carried out on Mueller Hinton Agar by Kirby Bauer method.

Results: The prevalence of uropathogen was found 35%. The Gram-negative bacteria were most common isolates in comparison to Gram- positive bacteria. E. coli was the most common isolates followed by S. saprophyticus. Gram-negative organism were found more sensitive to Amikacin, Piperacillin Tazobactum, Ampicillin sulbactum, Cefoperazone sulbactum and Nitrofurantoin. Gram-positive bacteria were found more sensitive to Piperacillin Tazobactum, Tetracycline, Ampicillin sulbactum and Linezolid. E. coli were found more sensitive to Amikacin, Ampicillin sulbactum, Piperacillin Tazobactum and Cefoperazone sulbactum.

Conclusion: Changing antimicrobial resistance pose challenge in treating urinary tract infections. Appropriate and judicious selection of antibiotic would limit the emerging drug resistant isolate in the future to treat this clinical condition successfully.

Keywords: Urinary Tract Infection, UTI, Uropathogens, reproductive age group females. Pregnant women, Antibiotic resistance, E. coli

Introduction

Urinary Tract Infections (UTI) frequently occur in both community and hospital environment are of the most common bacterial infections in humans. The outcomes of urinary tract infections are increase hospitalization, increase patient treatment cost and mortality.[1] Bacteria can invade and cause UTI via two major routes ascending and heamatogenous pathway. Ascending route is most common route of infection in females. Urinary tract infections are characterized as either upper or lower UTI primarily on the basis of anatomical location of infection. The lower UTI affecting the bladder and urethra and the upper UTI affecting the ureter and kidneys mainly.

Women are at three times greater risk for UTI then man because of short, straight anatomy of the urethra, and termination of female urethra beneath the labia resulting in colonization by colonic gram negative bacilli.[2] Most of the UTI are caused by gram-negative bacteria like E. coli, Proteus species, Klebsiella species, Pseudomonas aeruginosa, Acinetobacter, Serratia and Morganella morganii. UTI also caused by gram positive bacteria like Enterococcus, Staphylococcus specially coagulase negative Staphylococci and Streptococcus agalactiae.[3]

E. coli are one of the most prevalent pathogens among gram-negative bacteria capable of causing complicated and uncomplicated UTI.[4] UTI during pregnancy leads to low birth weight babies, increase perinatal mortality and premature births along with acute and chronic sequelae in mothers.[5] Diagnosis and definitive treatment of UTI mainly based on bacteriological culture and antibiotic sensitivity. In past Decade, indiscriminate use of antibiotics resulted in word wide rise of multidrug resistance cases.[6]

Hence, present study planned to find out the prevalence of urinary pathogens and their antibiotic resistance pattern in reproductive age group females to provide better cost effective treatment to female patients.

Aim-: To Detect the micro-organism & to make people aware for the causes of uropathogens

Review & literature

The Cochrane Review by Villar et al assessed the effects of different durations of treatment for asymptomatic bacteriuria and concluded that there was insufficient evidence to evaluate whether a single dose or longer-duration doses were equivalent in treating asymptomatic bacteriuria. Ten studies were included that compared single dose treatment with 4-7 day treatment. The risk of failing to cure asymptomatic bacteriuria was higher for 1 day treatment than for 7 days of treatment although the difference was not statistically significant.

Historically, ampicillin has been the drug of choice, but in recent years E.coli has become increasingly resistant to ampicillin. Ampicillin resistant found in 20 to 30% of E.coli cultured from urine in the outpatient setting. Currently, 30-50% of E.coli are

ampicillin resistant, and 20-30% are cephalosporin resistant.

Fosfomycin trometamol which is a derivative of phosphonic acid is a new antibiotic that can be taken as a single dose 3g sachet orally which is equally effective as 7-10 day course of nitrofurantoin, norfloxacin or cotrimoxazole. This drug is active against E.coli, enterococci and Citrobacter, Enterobacter, Klebsiella and Serratia species .

The incidence of extended spectrum β lactamase (ESBL) producing strains among clinical isolate has been steadily increasing over the past few years, resulting in the limitation of therapeutic options. Microorganisms responsible for urinary tract infection (UTI), especially Escherichia coli and klebsiella spp. have the ability to produce ESBLs in large quantities. These enzyme are encoded by transferable conjugative plasmids, which often code resistance to cephalosporins as well as to other antibiotics. The most frequent co-resistances found in ESBL producing organisms are to aminoglycosides, fluoroquinolones, tetracyclines, and sulfamethoxazole- trimethroprim .

There is a rising incidence of urinary tract infection (UTI) with ESBL producing bacteria. In hospitalized patients with positive cultures for ESBL producing Escherichia coli, the majority of the isolates are attributed to a clinical infection rather than colonization. The commonest clinical specimen to yield the organism was urine, which was positive in 57.8% of patients .

Supriya S.et al in their study found that of the 217 isolated, 87 were cephotaxime resistant Gram-negative bacilli. Of these, 42 (48.3%) were found to be ESBL producers. Escherichia coli, Klebsiella pneumoniae and Acinetobacter were ESBL producing species.

In India antimicrobial susceptibility pattern of uropathogens vary widely by region. High resistance rates to oral antibiotics have been observed, probably due to uncontrolled consumption of these antibiotics. Resistance to amikacin, piperacillin tazobactum and meropenem are low, likely reflecting lower usage of these drugs.

In 2002, SV Lavanya et al , in their study reported that 35.7% of cases were sensitive to Cephalexin, 28.5% to Nitrofurantoin, 23.8% to Amoxicillin and 11.9% to Norfloxacin.

Aziz Marjhan et al From Pakistan in 2006 stated that Escherichia coli showed 66.67% resistance to ampicillins and sulphonamides. Enterobacters showed 100% resistance to ampicillins, cephalosporins and nitrofurantoin. Staphylococcus saprophyticus showed 66.67% resistance to ampicillins and sulphonamides .

R J Girishbabu from Tumkur in 2011 found that Pipercillin-Tazobactum, amikacin and nitrofurantoin were found to be the most effective antibiotics against the urinary isolates. Akinola B. Ajayi from Nigeria found that Staphylococcus aureus, the commonest isolate showed good sensitivity to gentamicin and nitrofurantoin with 75% and 77.8% of the organisms respectively. Proteus spp showed good sensitivity to gentamicin, nalidixic acid, ceftazidime and cefuroxime. Most of the organisms showed good sensitivity to nitrofurantoin and gentamicin.

C. Obirikorang in Ghana in 2012 also showed that most of the E. coli isolated were sensitive to nitrofurantoin and gentamicin

Materials and Methods

A cross sectional study was carried out in the department of microbiology,Jayoti Vidyapeeth Women's University, Jaipur from January 2020 to September 2020. A total no.of 105 urine samples were collected from female patients clinically suspected of urinary tract infection. The women in reproductive age group 16 to 45 years with history of urinary tract infection were included in the study and only one sample was collected from each patient. The exclusion criteria were leaky or dirty container, delay in transportation of sample more than 2 hours, previous history of antibiotics, surgery or operative procedure. Midstream clean catch urine samples were collected and transported to microbiology laboratory for processing. Urine culture and antibiotic susceptibility testing was performed in laboratory.

The urine culture was done using a sterile calibrated loop of 4 mm diameter delivering 10 microliter volume of urine. A loopful of well mixed un- centrifuged urine was inoculated on the Mac-Conkey Agar and Blood Agar media plates. All plates were incubated at 37^0 C aerobically for 24 hours. The bacterial growth was identify by Gram stain, motility and a set of biochemical test including catalase, coagulase, oxidase, indole, methyl red, Voges Proskauer, citrate, urease and triple sugar iron medi Antimicrobial susceptibility testing was performed by using Kirby Bauer disc diffusion method as described by the national committee for clinical laboratory standard (presently called clinical laboratory standard institute).[8] Interpretation as sensitive or resistant was done on the basis of diameter of zone of inhibition of bacteria growth on Mueller Hinton Agar plate as recommended by Hi Media disc manufacturer. Antibiotic discs used for susceptibility testing for gram negative bacteria were Amikacin (30 µgm), Ampicillin sulbactam (10 / 10 µgm), Ceftriaxone (30 µgm), Cefotaxime

(30 µgm), Ciprofloxacin (5 µgm), Cefixime (5 µgm),

Ceftazidime (30 µgm), Cefoperazone sulbactum (75 / 30 µgm), Co-trimoxazole (25 µgm), Norfloxacin (10 µgm), Nitrofurantoin (300 µgm), Ofloxacin (5 µgm), Piperacillin tazobactam (100 / 10 µgm) and Imipenem (10 µgm). Antibiotic disc used for gram-positive bacteria includes Ceftazidime (30 µgm), Ciprofloxacin (5 µgm), Cloxacillin (100 µgm), Co-trimoxazole (25 µgm), Gentamicin (10 µgm), Norfloxacin (10 µgm), Nitrofurantoin (300 µgm), Penicillin (10 U), Piperacillin tazobactam (100 / 10 µgm), Tetracycline (30 µgm), Vancomycin (30 µgm) and Linezolid (30 µgm). Data collected and analysis was done using appropriate statistical methods.

Results and Discussions

In present study, the frequency of uropathogens in reproductive age group female was found to be 36% (Table 1). B. Shanthi et al. (2018)[9] was found the higher percentage due to their large sample size. The maximum number of cases was found in the age group 16 – 30 years and minimum number of cases was found in the age group 31 – 45 years (Table 2). The reason being women in this age group are more sexually active and more prone to develop UTI probably due to characteristic anatomy of the urethra and the effect of normal physiological changes that affects the urinary tract – short urethra, its close proximity to the anus, urethral trauma during intercourse, dilatation of urethra and stasis of urine during pregnancy.[10,11]

Out of 105 cases, 13 females were found pregnant and 92 were found non-pregnant. All 13 pregnant women were found culture positive due to a number of factors including urethral dilation, increase bladder volume and decrease bladder tone, along with decrease urethral tone, which contributes to increase urinary stasis and vesicoureteral reflex and up to 70% of pregnant women develop glycosuria, which favors bacterial growth in the urine.[12]

Gram-negative bacteria dominated over gram-positive bacteria as the etiological agent for UTI as shown in Table 3.

E. coli was the most common isolates both in non-pregnant and pregnant women followed by Staphylococcus saprophyticus. The other micro-organisms isolated were Staphylococcus aureus, Klebsiella pneumoniae, Proteus mirabilis, Citrobacter freundii, Enterococcus faecalis. Out of total isolates, 52% were E. coli and 17.4% were Staphylococcus saprophyticus. Similar results were foundby Goyal Ankur et al. 2015[13] Agersew Alemu et al. 2012[14] and Geeta Gupta et al. 2019.[15]

The gram-negative bacteria were found more sensitive to antibiotics Amikacin, Piperacillin Tazobactum, Ampicillin sulbactum, Cefoperazone sulbactum and Nitrofurantoin (Table 4). Similar results were found by B. Shanthi et al. (2018)[9] and Obiogbolu et al. (2009).[16] The gram-positive bacteria were found more sensitive to antibiotics Piperacillin Tazobactum, Tetracycline, Ampicillin sulbactum and Linezolid (Table 5). The most effective antibiotic for the E. coli was found to be Amikacin and Ampicillin sulbactum, Iram Shaifali et al. (2012)[17] observe Nitrofurantoin followed by Amoxicillin, Nalidixic Acid and Co-Trimoxazole were sensitive. The most effective antibiotic for the S. saprophyticus was Tetracycline and Levofloxacin. Adedeji BA et al. (2009)[18] found Gentamycin and Ofloxacin were the most active antibiotics and isolates showed high resistance to Co-Trimoxazole and Amoxicillin, The reason behind different antibiotic susceptibility pattern of isolates from other studies because sensitivity varies widely by region, OPD and IPD patients included in the study.

Table 1: Showing growth pattern among pregnant and non-pregnant women

S. No.	Growth	Pregnant	Non-Pregnant	Total
1	Present	13	28	41
2	Absent	-	64	64
Total		13	92	105

Table 2: Showing age group wise distribution of pregnant and non-pregnant women

S. No.	Age Group (Yrs)	Pregnant	Non-Pregnant	Total
1	16 – 30	13	29	42
2	31 – 40	-	35	35
3	41 – 45	-	28	28
Total		13	92	105

Table 3: Distribution of isolated micro-organisms in culture

S. No.	Micro-organism	No. of Women		Total
		Pregnant	Non-Pregnant	
1	E. coli	06	15	21
2	S. saprophyticus	04	03	07
3	S. aureus	--	04	04
4	Proteus species	--	02	02
5	Citrobacter freundii	--	02	02
6	Enterococcus	--	01	01
7	Klebsiella pneumoniae	02	01	03
8	CONS	01	--	01
9	No Organism Isolated	--	64	64
Total		13	92	105

Table 4: Antibiotic susceptibility pattern of Gram-negative organisms. (N=23)

S. No.	Antibiotic	Sensitive N (%)	Intermediate N (%)	Resistant N (%)
1	Ampicillin	13 (56.52)	0 (0)	6 (26.08)
2	Piperacillin	13 (56.52)	1 (4.34)	0 (0)
3	Ceftriaxone	2 (8.69)	1 (4.34)	0 (0)
4	Ceftazidime	5 (21.73)	0 (0)	9 (39.13)
5	Tetracycline	7 (30.43)	0 (0)	10 (43.42)
6	Nitrofurantoin	8 (34.78)	2 (8.69)	2 (8.69)
7	Ciprofloxacin	1 (4.34)	1 (4.34)	4 (17.30)
8	Amikacin	16 (69.56)	1 (4.34)	0 (0)
9	Levofloxacin	3 (13.04)	2 (8.69)	4 (17.30)
10	Co-Trimoxazole	6 (26.08)	0 (0)	5 (21.73)
11	Meropenem	2 (8.69)	1 (4.34)	4 (17.30)
12	Imipenem	3 (13.04)	1 (4.34)	0 (0)
13	Cefoperazone sulbactum	12 (52.17)	2 (8.69)	0 (0)

Table 5: Antibiotic susceptibility pattern of Gram-Positive organisms. (N=13)

S. No.	Antibiotic	Sensitive N (%)	Intermediate N (%)	Resistant N (%)
1	Amoxicillin	2 (15.38)	0 (0)	0 (0)
2	Co-Trimoxazole	5 (38.46)	2 (15.38)	3 (23.07)
3	Ofloxacin	4 (30.76)	0 (0)	1 (7.69)
4	Linezolid	9 (69.23)	0 (0)	0 (0)
5	Vancomycin	2 (15.38)	1 (7.69)	0 (0)
6	Ampicillin sulbactum	10 (76.92)	0 (0)	1 (7.69)
7	Tetracycline	11 (84.61	0 (0)	1 (7.69)
8	Levofloxacin	9 (69.23)	0 (0)	0 (0)
9	Piperacillin Tazobactum	1 (7.69)	0 (0)	0 (0)
10	Ciprofloxacin	9 (69.23)	1 (7.69)	1 (7.69)

Conclusion

The present study raised awareness regarding high vulnerability of women in reproductive age group for urinary tract infections. The pregnant women were found more prone to develop UTI in comparison to the non-pregnant women. The study provide information regarding uropathogens and their antibiotic susceptibility pattern. Gram-negative bacteria were found more common isolates in comparison to gram positive bacteria causing UTI. Analyzing antibiotic susceptibility pattern of uropathogens will help to overcome the therapeutic dilemmas and to guide in selection of appropriate antibiotics for empirical treatment to the patients**Funding:** Nil.

Conflict of Interest: None declared.

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