 DOI: 10.18231/2455-6807.2017.0019

Antibiotic resistance pattern of urinary isolates in a Rural Medical College of Maharashtra

Sanjaykumar More[1], Sourabh Chakraborty[2,*], Sandeep L. Nilekar[3], Deepali M. Kulkarni[4], Rajesh S. Ovhal[5]

[1]Professor & HOD, [2]Resident, [3,4]Associate Professor, [5]Assistant Professor, Dept. of Microbiology,

***Corresponding Author:**
Email: dr.sourabhchakraborty@yahoo.in

Abstract

Objective: To determine resistance pattern of urinary isolates in a rural medical college of Maharashtra.

Methods: Urine samples were collected using midstream clean catch method in clinically diagnosed UTI cases from august 2016 to February 2017. A total of 320 samples were analyzed under supervision of qualified microbiologist. Antibiotic sensitivity of isolated microorganisms were tested for commonly used antibiotics by Kirby bauer technique.

Results: Out of 320 samples 200 were positive. The pathogens isolated were E.coli 60 (30%), Pseudomonas 45 (22.5%), Methicillin resistant Staphylococcus aureus 35 (17.5%) & Klebsiella 30 (15%). E.coli being the most common isolate. The isolates showed high degree resistance for Amoxycillin & Cotrimoxazole.

Conclusion: 62.5% of samples showed significant growth. Moderate yield suggest moderate clinical correlations in suspected cases of UTI. E.coli showed low level resistance to Nitrofurantoin (1.67%), Amikacin (3.34%), Norfloxacin (13.3%) & high degree resistance to Cotrimoxazole (63.34%), Piperacillin + Tazobactam (50%), fluoroquinolones (27.04%). It's very clear from the study E.coli is the MOC infecting uropathogen & developing resistance against the commonly used antibiotics.

Keywords: Uropathogens, Antibiotic resistance, Urinary tract infections.

Introduction

Urinary tract infection is not only common nosocomial infection but an important source of morbidity in community as well.[1,2] It is the most frequent cause of illness in human after respiratory infections.[3] Neonates, girls, young women and older men are most susceptible to UTI .[4]

Urinalysis only indicates presence of bacteria and leucocytes in the urine, which is indirect evidence of UTI but it can only be confirmed on the basis of microscopy and microbial culture.[5] In most instances, growth of more than 10^5 organisms per milliliter from a properly collected midstream "clean-catch" urine sample indicates infection. However, significant bacteriuria is lacking in some cases of true UTI, especially in symptomatic patients, a smaller number of bacteria (10^2 to 10^4/mL) may signify infection. In urine specimens obtained by suprapubic aspiration or "in-and-out" catheterization and in samples from a patient with an indwelling catheter, colony counts of 10^2 to 10^4/mL generally indicate infection. Conversely, colony counts of $>10^5$ /mL of midstream urine are occasionally due to specimen contamination, which is especially likely when multiple species are found.[6]

The vast majority of uncomplicated UTIs are caused by the Gram-negative bacillus Escherichia coli, with other pathogens including Enterococci, Staphylococcus saprophyticus, Klebsiella spp. and Proteus mirabilis.[7] The extensive and inappropriate use of antimicrobial agents has invariably resulted in the development of antibiotic resistance which, in recent years, has become a major problem worldwide.[8] Various virulence factors in uropathogens including adhesions, hemolysins, capsular polysaccharides and drug resistance are major decisive factors in development and treatment of infections.[9]

In patients with suspected UTI, antibiotic treatment is usually started empirically, before urine culture results are available. To ensure appropriate treatment, knowledge of the organisms that cause UTI and their antibiotic susceptibility is mandatory.[10] There are various reports available about changing pattern

of pathogen and their susceptibility to routinely used antibiotics in last two decades due to extra chromosomal genetic elements, which simultaneously carry gene for resistance to number of antibiotics and this has made the situation miserable, especially in gram negative bacteria.[11]

So the databank about antibiotic resistance has to be updated regularly, to achieve maximal clinical response before the sensitivity report gets available and also extensive and inappropriate use of antibiotics can be avoided.

The study is aimed to determine pattern of pathogens isolated, their resistance and sensitivity pattern to different antibiotics in a rural govt. medical college of Maharashtra, so the clinicians can start more appropriate treatment empirically.

Materials and Method

This study was conducted in department of Microbiology, in a rural govt. medical college & hospital of Maharashtra from august 2016 to February 2017. A total of 320 samples were analyzed of patients who were suspected to be having UTI, with prior permission from Institutional Ethics Committee. Samples were collected from clinically diagnosed UTI cases (fever, dysuria, increased frequency of urination).

Specimen were collected by standard "clean catch" mid stream method in patients who had no catheter in place, while in catheterized patients, sample was collected in sterile, screw capped, wide mouth container after clamping the catheter for 30 mins. Before collecting the sample, male subjects were asked to clean the genital parts with soap and water and in case of female, they were asked to wash vulva & to separate carefully the labia prior to voiding the urine in sterile bottle. Samples were examined and processed on the Blood agar and Mckonkey's agar medium by standard loop method and incubated for atleast 24 hours at 37degree. Plates were observed for bacterial growth.[12]

Uropathogens were identified on the basis of Gram's reaction, colony morphology and standard biochemical tests. Antibiotic susceptibility test was carried out for bacterial isolates by Kirby Baur disc diffusion technique. Antibiotics against which sensitivity was tested in the present study included Amoxycillin, Amoxiclav, Ciprofloxacin, Norfloxacin, Cotrimoxazole, Gentamicin, Amikacin, Nitrofurantoin, Ceftazidime, Meropenem, Piperacillin + Tazobactam.[13]

Table 1: Antibiotic Resistance and Ssensitivity Pattern of Isolated Uropathogens

	E.coli (60)		**K.pneumoniae (30)**		**P.aeruginosa (45)**		**MRSA (35)**		**C.freundii (15)**		**C.koserii (10)**		**Enterobacter (5)**		**Mean**	
	S	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**	**R**
AMX	13 (21.66)	47 (78.34)	6 (20)	24 (80)	**ND**	**ND**	18 (51.42)	17 (48.58)	7 (46.67)	8 (53.33)	4 (40)	6 (60)	1 (20)	4 (80)	**33.30**	**66.70**
AMX + Clav	19 (31.67)	41 (68.33)	19 (63.33)	11 (36.67)	**ND**	**ND**	26 (74.28)	9 (25.72)	8 (53.33)	7 (46.67)	6 (60)	4 (40)	2 (40)	3 (60)	**53.77**	**46.23**
COT	22 (36.67)	38 (63.33)	5 (16.67)	25 (83.33)	**ND**	**ND**	19 (54.29)	16 (45.71)	6 (40)	9 (60)	4 (40)	6 (60)	2 (40)	3 (60)	**37.94**	**62.06**
GEN	45 (75)	15 (25)	20 (66.67)	10 (33.33)	26 (57.78)	19 (42.22)	23 (65.71)	12 (34.29)	9 (60)	6 (40)	7 (70)	3 (30)	3 (60)	2 (40)	**65.02**	**34.98**
AK	57 (96.66)	3 (3.34)	26 (86.67)	4 (13.33)	31 (68.89)	14 (31.11)	**ND**	**ND**	12 (80)	3 (20)	8 (80)	2 (20)	4 (80)	1 (20)	**82.03**	**17.97**
CIP	25 (41.67)	35 (58.33)	21 (70)	9 (30)	36 (80)	9 (20)	28 (80)	7 (20)	11 (73.3)	4 (26.67)	7 (70)	3 (30)	2 (40)	3 (60)	**65**	**35**
NX	52 (86.67)	8 (13.33)	25 (83.33)	5 (16.67)	38 (84.44)	7 (15.56)	24 (68.57)	11 (31.43)	12 (80)	3 (20)	8 (80)	2 (20)	4 (80)	1 (20)	**80.43**	**19.57**
NIT	59 (98.33)	1 (1.67)	23 (76.67)	7 (23.33)	**ND**	**ND**	34 (97.14)	1 (2.86)	13 (86.67)	2 (13.33)	8 (80)	2 (20)	4 (80)	1 (20)	**86.46**	**13.54**
CAZ	44 (73.33)	16 (26.67)	12 (40)	18 (60)	40 (88.89)	5 (11.11)	**ND**	**ND**	8 (53.33)	7 (46.67)	4 (40)	6 (60)	2 (40)	3 (60)	**55.93**	**44.07**
MRP	56 (93.33)	4 (6.67)	28 (93.33)	12 (6.67)	39 (86.67)	6 (13.33)	**ND**	**ND**	13 (86.67)	2 (13.33)	9 (90)	1 (10)	3 (60)	2 (40)	**85**	**15**
PIT	30 (50)	30 (50)	22 (73.33)	8 (26.67)	36 (80)	9 (20)	**ND**	**ND**	11 (73.33)	4 (26.67)	14 (93.33)	1 (6.67)	5 (100)	0 (0)	**78.33**	**21.67**

Result

A total of 320 urine Samples were analyzed in the present study. Out of 320 samples, 200 samples were positive. Most common organism isolated were gram negative organisms. E.coli (30%), Pseudomonas (22.5%), followed by MRSA (17.5%) and Klebsiella (15%). The antibiotics were chosen as per the CLSI guidelines. E.coli showed low level resistance for Nitrofurantoin (1.67%), Amikacin (3.34%), Norfloxacin (13.33%) with high resistance pattern for Cotrimoxazole (63.33%), Fluoroquinolones (35.83%), Piperacilin & Tazobactam (50%). Pseudomonas showed low resistance for Ceftazidime (11.11%), Meropenem (13.33%), Norfloxacin (15.56%). MRSA was highly resistant to Cotrimoxazole (45.71%), Amoxycillin (48.58%), whereas excellent sensitivity to Nitrofurantoin (97.14%). Klebsiella was less resistant for meropenem (6.67%), Norfloxacin (16.67%), Amikacin (13.33%), whereas highly resistant for Cotrimoxazole (83.33%), Amoxycillin (80%), Ceftazidime (60%).

The isolates showed high degree of resistance for Amoxycillin and Cotrimoxzole (Table 1).

Discussion

UTI is a major health problem worldwide & pattern of antibiotic resistance varies in different regions. Injudicial usage of higher antibiotics at community level, making the situation more alarming. In community and hospital setting the etiology of UTI and antimicrobial susceptibility of bacteria causing UTI have been changing over the years.[14,15]

Looking at the result the most common organism isolated in our hospital is E.coli (30%), this is consistent with other studies Stamm WE et.al (2002), Biswas D et.al (2006) in which E.coli is the most common pathogen isolated from patients with UTI.[6,16] Following E.coli our study shows Pseudomonas (22.5%), MRSA (17.5%), Klebsiella (15%) as the common isolates. Our study is in contrast with a study by Jai Pal Paryani et.al (2012) showing klebsiella (11.31%), Enterobacter (11.31%), Proteus (7.86%) as the common isolates other than E.coli (64.41%)[17] or a study showing after E.coli (most common) klebsiella, pseudomonas and staphylococcus are the next common isolates.[18] Enterobacteriaceae having several factors for their attachment to urothelium. The Gram negative bacilli bacteria colonize the urogenital mucosa with adhesion, pilli, fimbriae, P1 blood group phenotype receptor.**[19]**

E.coli found to be resistant to the 1st line antibiotics like: Amoxycillin (78.34%), Amoxycillin & Clavulanic acid (68.33%), Cotrimoxazole (63.33%). This is in accordance with Srinivasa H et.al (1999) stating about resistance of microorganisms to conventional antibiotics.**[20]**

Pseudomonas aeuroginosa which is the most common cause of hospital acquired UTI, was less resistant to Fluroquinolones (17.78%) and cephalosporine (ceftazidime 11.1%) than Aminoglycoside (Amikacin 31.1%), in contrast to a study by Jai Pal Paryani al., 2012.**[17]**

MRSA is the 3rd most common isolate in our study, similar to studies by shalini et al., 2011**[18]** and molay Banerjee et al., 2014**[21]** where staph is the 4th most common isolate. For MRSA resistance pattern is much better than GNB's.

In our study mean resistance for Cotrimoxazole, amoxicillin & Clavulanic acid, Cephalosporine (ceftazidime), Quinolones (ciprofloxacin & Levofloxacin), were 62.06%, 46.23%, 44.07, 27.29 respectively. From this study it can be seen that Amoxicillin & Clavulanic acid and Cotrimoxazole are nearly having no role against uropathogens due to high degree of resistance. Least resistance pattern were shown by Nitrofurantoin (13.54%), Meropenem (15%), Norfloxacin (19.57%).

Our study suggest Nitrofurantion (13.54% resistance) and Norfloxacin (19.57% resistance) as the 1st line drug before culture and sensitivity report is available. Both are active against E.coli, klebsiella, MRSA. Both are cost effective and readily available. Nitrofurantoin is better in pregnancy than Norfloxacin.**[22]** Recent study in India showed Nitofurantoin having best in-vitro susceptibility profile against E.coli. The minimal resistance of Nitrofurantoin against E.coli (1.67%) may be influenced by Nitrofurantoin's narrow distribution and limited interaction with pathogens outside urinary tract.

In a recent study by Bours PH et.al (2010) 29.5% of E.coli was suspected to produce ESBL and, Amikacin and Nitrofurantoin were the only drugs to which > 90% of E.coli was susceptible.**[21]** In the present study Nitrofurantoin is resistant against 1.7% isolates of E.coli, 2.86% isolates of MRSA, 23.3% isolates of klebsiella compared to Norfloxacin resistant to 13.3%, 31.4%, 13.3% respectively but with an advantage of activity against pseudomonas (15.6% resistance).

Conclusion

In the present study of all samples 62.5% showed significant growth. Moderate yield of positive culture suggest not so good clinical correlation in suspected cases of UTI. But it is certain that due to the rising trend of antibiotic resistance, choice of antibiotics getting lesser day by day. Cotrimoxazole and Ampicillin which was drug of choice previously, now rarely prescribed as empirical therapy. But Norfloxacin and Nitrofurantoin are good alternative to start with antibiotic therapy for ESBL producing E.coli.

References

1. Sharma S. Current understanding of pathogenic mechanisms in UTis. Ann Natl Acad Med Sci 1997;33:31.
2. Acharya A, Gautam R, Subedee L. Uropathogens and their antimicrobial susceptibility pattern in Bharatpur, Nepal. Nepal Med Coll J 2011;13:30-3.
3. Liperky BA. Urinary tract infection in men: Epidemiology, pathophysiology, diagnosis and treatment. Ann Intern Med 1989;111:138.
4. Nicole W, Jon DM. Deciphering Dysuria. Emerg Med. 2008;40(9):29.
5. Van Nostrand JD, Junkins AD, Bartholdi RK. Poor predictive analysis and microscopic examination to detect urinary tract infection. Am J Clin Pathol 2000;113:709.
6. Stamm WE, Schaeffer AJ. The State of the Art in the management of Urinary Tract Infections. Am J Med 2002; (suppl 1 A): 113.
7. Blondeau JM. Current issues in the management of urinary tract infections: extended-release ciprofloxacin as a novel treatment option. Drugs. 2004;64(6):611-28.
8. Goldstein FW. Antibiotic susceptibility of bacterial strains isolated from patients with community-acquired urinary tract infections in France. Multicentre Study Group. Eur J Clin Microbiol Infect Dis. 2000;19:112-7.
9. Seigfried L, Kmetova M, Puzova H, Molokacova M, Filkas J. Virulence associated factors in E. Coli strains isolated from children with UTI. J Med Microbiol 1994;41:127.
10. Ashkenazi S, EvenTov S, Samra Z, et al. Uropathogens of various childhood populations and their antibiotic susceptibility. Pediatr Infect Dis J. 1991;10:742–6.

11. Ram S, Gupta R, Gaheer M. Emerging antibiotic resistance among uropathogens. Ind J Med Sci 2000;54:388.
12. Tille, P. M. (2007). Infections of the Urinary tract. In *Bailey & scott's Diagnostic Microbiology* (pp. 923-927). china: Elsevier.
13. CLSI. *Performance Standards for Antimicrobial Susceptibility Testing.* 26th ed. CLSI supplement M100S. Wayne, PA: Clinical and Laboratory Standards Institute; 2016.
14. New HC. Urinary tract infections. Am J Med. 1996; 100 (Suppl.4A): S63-70.
15. Jones RN. Impact of changing pathogens and antimicrobial susceptibility pattern in treatment of serious infections in hospitalized patients. Am J Med. 1996; 100 (Suppl.6A): S3-12.
16. Biswas D, Gupta P, Prasad R, et al. Choice of antibiotic for empirical therapy of acute cystitis in a setting of high antimicrobial resistance. Indian J Med Sci. 2006;60:53-58.
17. Jai Pal Paryani, Shafique-ur-Rehman Memon, Zakir Hussain Rajpar, Syed Azhar Shah, "Pattern and Sensitivity of Microorganisms Causing Urinary Tract Infection at Teaching Hospital" JLUMHS May-August 2012; Vol 11: No. 02.
18. Shalini, Joshi MC, Rashid MK, Joshi HS, "Study of Antibiotic Sensitivity Pattern In Urinary Tract Infections At A Tertiary Hospital" NJIRM 2011; Vol. 2(3). July-September.
19. Das R, Chandrasekhar TS, Joshi HS, Gurung M, Shreshtha N, Shivananda PG. Frequency and susceptibility profile of pathogens causing urinary tract infections at a tertiary care hospital in western Nepal. Singapore Med J 2006;474:281–5.
20. Srinivasa H, Parija SC, Bhattacharya S, Sehgal R. Incidence of ciprofloxacin resistance in urinary isolates in eastern Nepal. J Comm Dis 1999;31:45.
21. Dr. Molay Banerjee1, Dr. Abhishek Arun, Dr. Sandeep Kr. Gupta, " Study of Culture and Sensitivity pattern of microorganisms in urinary tract infection at a tertiary care hospital" International Journal of Advanced Research (2014), Volume 2, Issue 11, 1000-1004.
22. Delzell JE Jr, Leferre ML. Urinary tract infections during pregnancy. Am Fam Physician. 2000;61(3):713-721.
23. Bours PH, Polak R, Hoepelman AI, et al. Increasing resistance in community-acquired urinary tract infections in Latin America, five years after the implementation of national therapeutic guidelines. Int J Infect Dis. 2010;14(9):770-4.