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Original Article

ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF UROPATHOGENS AT A TERTIARY CARE HOSPITAL IN CENTRAL INDIA DURING COVID ERA.

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ABSTRACT

Objective: The purpose of this study was to study the bacteriological profile of UTI in patients attending the tertiary care hospital and to study the antimicrobial sensitivity pattern of uropathogens.

Methods: This cross-sectional study was conducted after obtaining clearance from the institutional ethics committee. Clean-catch mid-stream urine samples were collected from patients symptomatic of UTIs. Samples were cultured aerobically on CLED agar. Isolates having significant growth (>10^5CFU/ml) were further processed for identification using standard microbiological techniques and their antimicrobial susceptibility pattern was evaluated by the Standard Kirby Bauer disk diffusion method as per CLSI 2020 guidelines.

Results: A total of 480 urine samples were processed, yielding 174 isolates. *Escherichia coli* (42.50%) was predominant, followed by *Klebsiella pneumonia*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Acinetobacter spp*., *Proteus spp.*, *Providencia spp.*, *Enterococcus spp.*, *Citrobacter spp.* and *Morganella morganii*. Gram-positive isolates exhibited high sensitivity towards vancomycin, linezolid, meropenem, and piperacillin tazobactum. Enteric coliforms exhibited high sensitivity towards colistin, meropenem, aminoglycosides, and piperacillin tazobactum. Non-fermenters exhibited high sensitivity towards colistin, meropenem, cefepime, and amoxycillin clavulanate.

Conclusion: The rampant injudicious irrational overuse of antibiotics has led to the emergence of multi-drug resistant bugs, which is posing a serious challenge to clinicians in the management of infections. Developing therapeutic protocols guided by susceptibility profiles for tuning antibiotic therapy regimens is an important strategy in tackling this menace.

Keywords: Urinary tract infection, Antimicrobial susceptibility, Antibiotic policy, Multi-drug-resistance, Empirical therapy, Isolates

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INTRODUCTION

Urinary tract infections (UTIs) are the most common cause of bacterial infections in humans. accounting for 25% of all infections. It is one of the most important causes of morbidity and also the second most common cause of hospital visits [1]. UTIs are defined by the presence of growth of more than 10^5 colony-forming units (CFU) of bacteria per ml of urine for asymptomatic individuals and much lower for symptomatic individuals (~10^3 CFU/ml) [2].

There are region-wise and time-wise variations in the susceptibility trends of Microorganisms causing UTIs [3].

It encompasses a wide array of infections, accounting for the community as well as hospital-acquired infections in developing countries. It is also the most common infectious disease in a clinical setting [1]. This problem spans all age groups, beginning from neonates to the geriatric age group [2].

The data of the past few years reflect that UTIs were the cause of 1 million visits to the Emergency Departments, seven million visits to the outpatient department, and about 100,000 cases of hospitalizations all over the world. About 150 million people suffer from UTI worldwide annually [4].

In almost all cases, there is a need to start treatment before the final microbiological results are available, which may lead to antibiotic resistance. Due to the rampant injudicious irrational use of antibiotics in infectious diseases and the lack of standardization in antimicrobial susceptibility tests, resistance to the commonly used antimicrobial agent is increasing year by year. This emerging trend is a global health threat and poses a serious challenge for clinicians in the management of such multi-drug-resistant infections. To aid better decision-making, the physician must have current knowledge of the prevailing pathogens and their antibiogram. Prompt diagnosis, culture report, and timely antimicrobial treatment help to minimize renal scarring and progressive kidney damage. This also plays a major role in preventing an uncomplicated UTI from going into a complicated one [3].

Regional data regarding the common uropathogens and their sensitivity pattern is required to guide the clinicians to start empirical therapy and is also beneficial in planning treatment protocols while managing UTIs. These data may be used to determine trends in antimicrobial susceptibilities, formulate local antibiotic policies, compare local with national data, and overall assist clinicians in the rational choice of antibiotic therapy to prevent misuse or overuse of antibiotics through antimicrobial stewardship.

In light of the above facts, and due to the paucity of such data from our region, we have planned a laboratory-based cross-sectional study to explore the bacteriological profile and antimicrobial susceptibility pattern of the Urinary tract infection cases treated in our clinical setup.

MATERIALS AND METHODS

This cross-sectional study was conducted in Dept. of Microbiology of a tertiary care hospital in Northern Madhya Pradesh from April 2021 to March 2022 after obtaining clearance from Institutional Ethics Committee. (Approval letter No.006/MIC/IECHP/DMC)

Study type: Cross-sectional study

Type of sampling: Convenience sampling

Sample size: 480

Duration of study: 1 year

Inclusion criteria: All the urine samples collected for culture from the suspected cases of UTI attending the tertiary care hospital including all OPD and inpatients, irrespective of their age and gender presenting with symptoms of UTI (burning micturition, fever, hematuria, dysuria, etc.) as a part of the routine diagnostic workup.

Exclusion criteria: The samples which on culture yielded>3 types of colonies suggestive of contamination and hence rendered inappropriate for further processing.

Specimen collection: All Clean-catch mid-stream urine samples were collected from patients symptomatic of UTI following a standard protocol to prevent contamination by normal vaginal, perineal, and anterior urethral flora for the consideration of a clinically relevant urine specimen.

Specimen transport: Urine specimens were being transported to the laboratory within the time limit so that they would undergo plating within 2 h. after collection; otherwise, refrigerated.

Isolation, identification, and characterization of organism: Samples were cultured aerobically on CLED agar, and strains having significant growth (>10^5cfu/ml) were further processed for identification using standard microbiological techniques**.** The samples yielding more than 3 types of colonies were rejected.

The antibiotic susceptibility test was done by the Kirby Bauer disc diffusion method by following standard procedures as per CLSI guidelines.

Data analysis: All data were maintained in Microsoft Office Excel and appropriate statistical tools were used wherever required [5-11].

RESULTS

A total of 480 urine samples were processed during the study period, out of which 174 (36.25%) samples were found to have significant bacterial growth.

Out of 174 culture-positive urine samples, 99 samples (56.89%) were obtained from females, while 75 were from male patients. The percentage of urinary tract infections was more in females as compared to males (table 1).

Table 1: Demographic profile of UTI cases

S. No.	Age group	No. of cases	Percentage
1.	0-20	13	7.47
2.	21-40	51	29.31
3.	41-60	62	35.63
4.	>60	48	27.58
	Gender		
1.	Male	75	43.10
2.	Female	99	56.89
	Total	174	36.25

As depicted in table 1, the prevalence of urinary tract infection was higher in the age group of 41-60 y (35.63%) followed by 21-40 y (29.31%).

In this study, both gram-negative and gram-positive organisms contributed to urinary tract infections in the study subjects. Out of the total 174 culture isolates, *Escherichia coli* (42.50%) was predominant, followed by *Klebsiella pneumoniae* (30.45%), *Pseudomonas aeruginosa* (14.94%), *Staphylococcus aureus* (5.17%), *Acinetobacter spp* (1.72), *Proteus spp.* (1.15 %), *Providencia spp. and Enterococcus spp* (1.14%), *Citrobacter spp.,* and *Morganella morganii* (0.57%) (fig. 1).

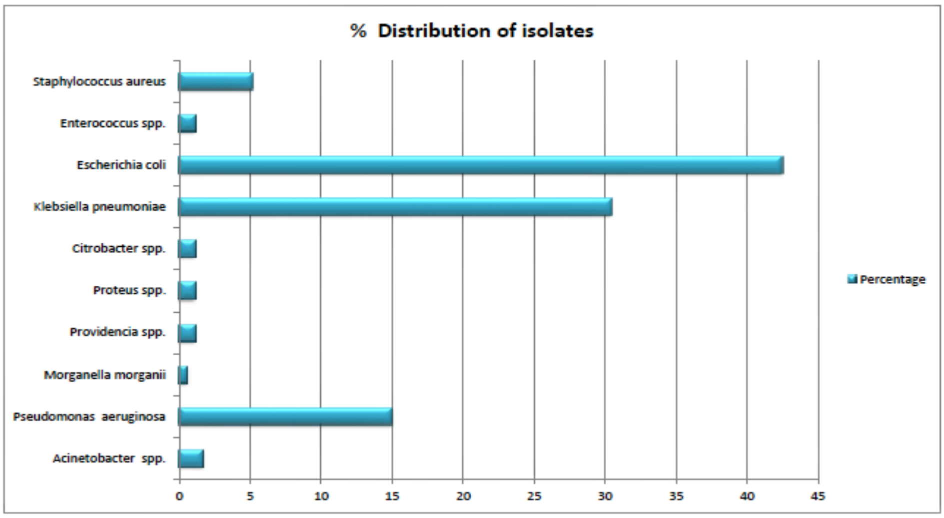

Fig. 1: Organism-wise distribution of urine culture isolates

In our study, gram-positive isolates exhibited high sensitivity towards vancomycin and linezolid, followed by meropenem, piperacillin tazobactum, Tetracycline, and levofloxacin. Amongst gram-negative isolates, enteric coliforms exhibited high sensitivity towards colistin, meropenem, aminoglycosides, and piperacillin tazobactum. Non-fermenters exhibited high sensitivity towards colistin, meropenem, cefepime, and amoxycillin clavulanate.

While studying the Antibiotic sensitivity pattern among gram-positive bacteria, *S. aureus* and *Enterococcus spp.* isolates were found to exhibit high sensitivity towards vancomycin (100%) and linezolid (89%), followed by meropenem, piperacillin-tazobactam, tetracycline, levofloxacin, amikacin, and clindamycin (table 2).

Regarding the antibiotic sensitivity pattern of enterobacteriaceae isolates, the most common isolate *Escherichia coli* showed high sensitivity towards colistin, meropenem, aminoglycosides, levofloxacin, and amoxycillin clavulanate. *E. coli* isolates exhibited the least susceptibility towards cotrimoxazole, 3rd generation cephalosporins (3GC), and Norfloxacin.

Klebsiella pneumoniae isolates were highly sensitive towards colistin, followed by meropenem, gentamycin, cefoperazone sulbactam, and ceftazidime clavulanate and highly resistant towards nitrofurantoin, fluoroquinolones, 3GC, and piperacillin tazobactum.

The *Citrobacter spp.* showed high sensitivity towards Meropenem, piperacillin tazobactam and colistin but low sensitivity towards aminoglycosides, 3GC, Fluoroquinolones, and nitrofurantoin. The *Proteus spp.* isolates showed modest sensitivity towards levofloxacin, colistin, and cefotaxime. The *Providencia spp.* isolates were highly sensitive to norfloxacin, colistin. meropenem and ceftriaxone.

The only *Morganella morganii* isolate was sensitive to amikacin, gentamycin, meropenem, and levofloxacin but resistant to β lactam-β lactamase inhibitors (BL-BLI), ceftazidime, ceftriaxone, nitrofurantoin, and cotrimoxazole (table 3).

Amongst non-fermenters, *P. aeruginosa* isolates exhibited high sensitivity towards colistin, meropenem, 3GC, cefepime, and amoxycillin clavulanate. but were found to be least susceptible to nitrofurantoin, norfloxacin, and aztreonam. *Acinetobacter spp.* Isolates were found to exhibit modest sensitivity towards colistin, meropenem, aminoglycosides, amoxycillin clavulanate, and cefepime (table 4).

Table 2: Antibiotic susceptibility pattern of GPC isolates

S. No.	Antibiotic	Susceptibility pattern n (%)	
		***S. aureus* (9)**	***Enterococcus spp.* (2)**
1.	Amoxycillin clavulanate	6 (67)	1 (50)
2.	Azithromycin	3 (33)	0
3.	Erythromycin	5 (55.55)	-
4.	Clindamycin	7 (78)	-
5.	Amikacin	8 (89)	1 (50)
6.	Gentamicin (10)	5 (55.55)	-
7.	Gentamicin (High level)	-	2 (100)
8.	Ciprofloxacin (5)	7 (78)	1 (50)
9.	Levofloxacin	7 (78)	2 (100)
10.	Co-trimoxazole	3 (33)	-
11.	Tetracycline	8 (89)	1 (50)
12.	Doxycycline	6 (67)	1 (50)
13.	Linezolid	8 (89)	2 (100)
14.	Vancomycin	9 (100)	2 (100)
15.	Meropenem	7 (78)	2 (100)
16.	Piperacillin tazobactum	7 (78)	2 (100)

Table 3: Antibiotic susceptibility pattern of Enterobacteriaceae isolates

S. No.	Antibiotics	No. of susceptible isolates n (%)					
		***E. coli* (74)**	***K. pneumoniae* (53)**	***Citrobacter spp.*(2)**	***Proteus spp.* (2)**	***Providencia Spp.*(2)**	***Morganella morganii* (1)**
1.	Amikacin	58 (78.4)	32 (60.4)	1 (50)	1 (50)	1 (50)	1 (100)
2.	Gentamycin	47 (63.5)	38 (72)	1 (50)	1 (50)	1 (50)	1 (100)
3.	Amoxycillin clavulanate	55 (74)	32 (60.4)	-	2 (100)	1 (50)	0
4.	Ceftazidime	22 (30)	21 (39.6)	0	2 (100)	2 (100)	0
5.	Ceftazidime Clavulanate	37 (50)	35 (66)	-	-	-	-
6.	Cefotaxime	13(17.57)	13 (24.5)	1 (50)	2 (100)		1 (100)
7.	Cefotaxime Clavulanate	26 (35)	27 (51)	-	-	-	-
8.	Ceftriaxone	46 (62.2)	32 (60.4)	1 (50)	1 (50)	2 (100)	0
9.	Cotrimoxazole	7 (9.46)	11 (21)	0	0	0	0
10.	Ciprofloxacin	33 (44.6)	16 (30.2)	1 (50)	1 (50)	1 (50)	1 (100)
11.	Levofloxacin	56 (75.7)	22 (41.5)	1 (50)	2 (100)	2 (100)	1 (100)
12.	Tobramycin	59 (79.7)	20 (38)	-	1 (50)	-	-
13.	Meropenem	68 (92)	47 (88.7)	2 (100)	1 (50)	2 (100)	1 (100)
14.	Piperacillin tazobactam	41 (55.4)	22 (41.5)	2 (100)	1 (50)	2 (100)	1 (100)
15.	Colistin	74 (100)	50 (94)	2 (100)	2 (100)	2 (100)	1 (100)
16.	Nitrofurantoin	37 (50)	13 (24.5)	1 (50)	1 (50)	1 (50)	0
17.	Norfloxacin	15 (20)	8 (15.1)	1 (50)	1 (50)	2 (100)	1 (100)
18.	Cefoperazone-sulbactam	46 (62.2)	38 (71.7)	1 (50)	1 (50)	0	0

Table 4: Antibiotic susceptibility pattern of non-fermenter isolates

S. No.	Antibiotics	No. of susceptible isolates n (%)	
		P. aeruginosa (26)	*Acinetobacter spp.(3)*
1.	Amikacin	14 (54)	2 (67)
2.	Gentamicin	11 (42.3)	2 (67)
3.	Cefepime	20 (77)	2 (67)
4.	Ceftazidime	22 (84.6)	1 (33.33)
8.	Ceftriaxone	20 (77)	1 (33.33)
11.	Amoxycillin clavulanate	22 (84.6)	2 (67)
12.	Piperacillin tazobactum	12 (46)	2 (67)
13.	Ciprofloxacin	10 (38.5)	1 (33.33)
16.	Meropenem	22 (84.6)	2 (67)
17.	Aztreonam	8 (31)	0
18.	Colistin	26 (100)	3 (100)
19.	Tobramycin	16 (61.5)	1 (33.33)
21	Cefoperazone sulbactam	13 (50)	1 (33.33)
25	Nitrofurantoin	7 (27)	1 (33.33)
26	Norfloxacin	4 (15.4)	1 (33.33)

DISCUSSION

UTI is a common problem faced by clinicians in every part of the world. It is a major burden in health care services due to the high prevalence in community and hospitals caused by different pathogenic organisms. So, regular surveillance of antibiotic sensitivity is required at the local level for choosing appropriate antimicrobial therapy for the management of such patients.

In our study, the prevalence of UTI turned out to be 36.25%. Similar prevalence rates were reported by several studies conducted in North India such as a study by Agrawal R et al. in NCR Ghaziabad and by Nilofar S et al.i n Anand,Gujarat. [12, 13].

But higher prevalence rates were reported by some other studies done by Tantry *et al.* (67%) and by Prakash *et al.* (53%) [14, 15]. The difference in positivity rates may be due to the differences in the selection of media, growth technique, and local prevalence rate.

The gender distribution of UTI cases in our study depicted female preponderance (56.89%) which is following the similar studies done by Rajendran V *et al.* (68.63%), Bency JAT *et al.* (63.3%), SinghVP *et al.* (65%) and Abu-Shaqra Q *et al.* [16-19].

The prevalence of UTI in women was more than in males. This finding is in concordance with many similar studies like Mukherji T *et al.* This is due to the short urethra being closer to the anus in females [20].

In our study, the highest positivity rate for uropathogens was reported in the age group 41-60 y (35.63%), a nearly similar observation was made by Das RN *et al.* (31.4%), and other studies done by Banerjee *et al.*, Obiogbolu *et al.*, and Shahina *et al.* [21-24].

The most common isolate in our study was *E. coli* (42.5%) followed by *Klebsiella pneumoniae* (30.45%) and *Pseudomonas aeruginosa* (15%). This observation is comparable to many studies done by Agrawal R *et al.*, Patel HK *et al.*, ShahA *et al.*, Sharma *et al.*, and Sabra M *et al.* [12, 25-28].

In our study, Gram-positive isolates exhibited high sensitivity towards Vancomycin and Linezolid followed by Meropenem, Piperacillin tazobactum, Tetracycline, and Levofloxacin.

This finding is in accordance with several similar studies in the recent past by Bency JAT *et al.*, Madhu GN *et al.*, Ghadage DP *et al.*, and Dasani S *et al.* [17, 29-31].

Amongst Gram Negative isolates, enteric coliforms exhibited high sensitivity towards Colistin, Meropenem, Aminoglycosides, and Piperacillin tazobactum.

This finding is in accordance with similar Indian studies in the recent past [32-35].

In our, study *E. coli* isolates exhibited very high sensitivity towards Colistin(100%) which is comparable with studies done by Birhman *et al.* in Greater Noida(100%) and Shah D *et al.* (98.43%) [32, 33] followed by Meropenem (92%) which is also comparable with similar studies done across India [33, 35, 36]. Susceptibility to Amikacin (78.4%) is closely comparable to the studies done by Shah D *et al.* where the sensitivity was 81.64% and by Somashekara *et al.* (84%) and higher than a recent study done by Harshkumar *et al.* [25, 33, 34]. This finding suggests amikacin still holds good to treat UTI.

In our study, *E. coli* isolates exhibited very poor susceptibility towards Cotrimoxazole (20%). Likewise, several similar studies have reported similar susceptibility patterns of *E. coli* towards cotrimoxazole, varying from 15.15% to 52.3% [15, 33, 35].

The second most commonly isolated uropathogen in our study was *Klebsiella pneumoniae.* In our study, *Klebsiella pneumoniae* isolates exhibited very high sensitivity to colistin (94%); closely similar findings were observed by Shah *et al.*, Saha *et al.*, and Varghese *et al.* [33, 38, 39]. *Klebsiella pneumoniae* showed low sensitivity towards nitrofurantoin (24.5%), which is comparable with various other studies across India, which show susceptibility range varying from 38-67%. Due to the increased production of cabapenemases *Klebsiella spp.* shows a high resistance rate towards carbapenems [15, 33].

In the present study, Non-fermenters exhibited high sensitivity towards Colistin, Meropenem, Cefepime, and Amoxycillin Clavulanate and low sensitivity towards Ciprofloxacin, Aztreonam, Nitrofurantoin and Norfloxacin. Apart from this, Acinetobacter spp. also exhibited modest sensitivity towards Piperacillin tazobactum and Gentamicin but high resistance to 3GC, unlike *Pseudomonas aeruginosa* isolates. These findings are comparable to similar studies conducted by Rajendran V *et al.*, Agrawal R *et al.*, and Shah D *et al.* [12, 16, 26].

One alarming finding in our study was the high degree of resistance to third-generation cephalosporins among most of the uropathogens, probably due to their rampant injudicious usage in clinical practice. Gross disregard towards culture-guided therapy and poor compliance to antibiotic therapy i.e. not taking appropriate antibiotics in the prescribed dose and duration, are major causes of the emergence of Multi-Drug Resistant infections.

Some of the important limitations of this study are that the antibiotic susceptibility pattern of uropathogens s was derived by the Standard disc diffusion test with results not confirmed by MIC determination; and that it is a single-center study with a limited sample size.

CONCLUSION

The frequent rampant injudicious irrational overuse of antibiotics as over-the-counter drugs has led to the rapid emergence of multi-drug resistant bugs, which is a global threat and is posing a serious challenge to the clinicians in the management of infections and their complications. As most of the routine antibiotics are being rendered ineffective over the course of time with a slow pace of developing newer molecules, clinicians are left with very few therapeutic options in their arsenal. Antibiotic stewardship is, therefore, necessary to restrict the injudicious use of antibiotics, which along with the infection control measures, can help in tackling this problem by obviating the selection pressure.

The data generated by this study can be compared with other similar studies in the region to determine the current changing trend of antimicrobial susceptibility patterns of uropathogens in our region. This would help determine the empirical therapy of UTIs and formulate local antibiotic policies, thus guiding clinicians in the rational choice of antibiotics to curb the misuse or rather an overuse of antibiotics.

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AUTHORS CONTRIBUTIONS

Concept, design, and Laboratory work were done by Mehta A and Gupta HK. Literature search and data acquisition were done by Gupta HK and Tripathi K. Data analysis and interpretation were done by Mehta A and Gupta HK. The manuscript was prepared by Mehta A and Tripathi K. Manuscript Editing and review were done by Mehta A and Gupta HK. All Authors read and approved the final manuscript.

CONFLICT OF INTERESTS

Declared none

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