# Microbiological Profile and Antibiogram of Bacterial Isolates Causing Urinary Tract Infection in Tertiary Care Hospital.

<sup>1</sup>Dr. Durgesh Dasharath Mahajan, <sup>2</sup>Dr. Pragati Abhimanyu Bulle,

Assistant Professor, Dept of Medicine, Shri Vasantrao Naik Government Medical College, Yavatmal. Assistant Professor, Dept of Microbiology, Shri Vasantrao Naik Government Medical College, Yavatmal.

## Abstract:

Urinary tract infections (UTIs) are counted among the most common infections in humans. In spite of the availability and use of the antimicrobial drugs, UTIs caused by bacteria have been showing increasing trends. Patients diagnosed clinically as UTI during the study period (January 2015 to December 2015) in a Shri Vasantrao Naik Government Medical College and Hospital, Yavatmal (Maharashtra) were included in the study. Urine sample of these patients were tested for Culture and antibiotic susceptibility testing. A total of 472 urine samples were included in the study of that 368 from female patients and 104 from male patients. Out of 472 urine samples urine culture was positive in 200(42.37%) whereas in 272 urine culture was negative. Most common etiological organisms of UTI isolated were *Escherichia coli* 64(32%) followed by *Klebsiella species* 61(30.5%), *Staphylococcus aureus* 26(13%), *Coagulase negative staphylococci (CoNS)* 11(5.5), *Pseudomonas aeruginosa* 11(5.5%) *Proteus species* 7(3.5%), *Citrobacter species* 4(2%), *Enterococci species* 4(2%), Acinetobacter species 2(1%), *Streptococcus pyogenes* 1(0.5%). Most effective antibiotics were imipenem, nitrofurantoin, amikacin and piperacillin-tazobactum. For prevention of UTIs, implementation of strict infection control guidelines, effective hand washing and judicious use of antimicrobials is mandatory to prevent the emergence of drug resistance among uropathogens.

Keywords: Urinary tract infection, Antibiotic sensitivity, Gram Negative, Gram Positive.

# I. Introduction

Urinary Tract Infections (UTIs) are defined by the presence of a growth of more than  $10^5$  colony forming units (CFU) of bacteria per ml of urine for asymptomatic individual and much lower for symptomatic individual (~ $10^3$  CFU/ml).<sup>1</sup> In urine sample obtained by supra pubic aspiration or in-and-out catheterization and in samples from a patient with an indwelling catheter, colony count of  $10^2$ - $10^4$ /ml generally indicates infection.<sup>2</sup>

Urinary tract infection (UTI) is one of the most commonly occurring community acquired infections in developing countries owing to lack of sanitation and unhygienic toilet habits.<sup>3</sup> Urinary tract infections (UTI) are caused by pathogenic invasion of the urinary tract which leads to an inflammatory response of the uroepithelium. Proliferation of bacteria in the urinary tract is the cause of urinary tract infection.<sup>4</sup>

An estimate of patients suffering from UTI is around 150 million per annum across the globe which may rise to75% in the female population by the age of 24 years and 15-25% of this group may suffer from the relapse of this diseases.<sup>5</sup> UTI in men are less common. It has been observed that up to one-third of all women will experience UTI at some point during their lifetime. This finding has been attributed to three features that facilitate ascending infections into bladder, namely a short urethra, the proximity of urethra to anus and colonization of vagina by the fecal flora.<sup>6</sup> About 150 million people are diagnosed with UTI each year, costing the global economy in excess of 6 billion dollars.<sup>7</sup>

# II. Aims and objective

## The present study was undertaken with the following aims and objectives.

1) To study the etiological profile of UTI

2) To determine the antibiotic sensitivity pattern of the bacterial isolates.

This was a retrospective study conducted for a period of one year (January 2015 to December 2015) in a Shri Vasantrao Naik Government Medical College and Hospital, Yavatmal (Maharashtra).

Inclusion criteria – i) With fever (> $38^{\circ}$ C) and chills ii) Patient showing one or more of the following symptoms Burning micturition, increased frequency, urgency of urine, dysuria and pain lower abdomen / flank pain/ supra pubic pain.

Exclusion criteria - i) Asymptomatic patients. ii) Patients already on antibiotic treatment (duration of 5-7 days) iii) Age less than 12 years. iv)Patients on indwelling catheters. Informed consent was taken from all the subjects participated in the study after explaining the study protocol. About 30 ml of clean catch midstream urine sample were collected in 100ml sterile, dry, leak-proof container with instructions on how to collect a clean catch

midstream urine (MSU). Urine is an excellent culture media and bacteria will multiply if specimen is left at room temperature for any appreciable time. For this reason, urine specimen was transported to the laboratory immediately after obtaining and was processed within one hour or in case of delay they were refrigerated at  $4^{\circ}$ C(upto 24 hours), until culture can be performed.<sup>8</sup>

#### **Direct microscopy**

Wet mount examination was performed to look for the presence of pus cells, epithelial cells, red blood cells, crystals or microorganisms. Sample culture, Bacterial identification and Antibiotic susceptibility testing Urine specimen was inoculated on Blood agar and MacConkey agar and incubated in incubator at 37 <sup>0</sup> C. Various organisms were identified on the basis of colony morphology and standard biochemical tests.<sup>9</sup> The isolates were subjected to antimicrobial susceptibility testing by Kirby Bauer disk diffusion method as per CLSI guidelines 2014.<sup>10,11</sup>

The following antibiotics used were: Nitrofurantoin  $(300\mu g)$ , Amikacin  $(30\mu g)$ , Cotrimoxazole  $(25\mu g)$ , Gentamycin  $(10\mu g)$ , Ciprofloxacin  $(5\mu g)$ , Ceftazidime  $(30 \ \mu g)$ , Norfloxacin  $(10\mu g)$ , Ampicillin  $(10\mu g)$  and Imipenem  $(10\mu g)$ , Piperacillin-tazobactum  $(100/10 \ \mu g)$ .

#### **Observations and results :**

A total of 472 urine samples were included in the study of that 368 from female patients and 104 from male patients. Out of 472 urine samples urine culture was positive in 200(42.37%) whereas in 272 urine culture was negative. (Table 1)

<b>Table 1</b> : Frequency of culture positives in males and females.								
Total no. of	Total no. of		Female		Male			
samples	culture positives	Number	Positive	%	Number	Positive	%	
472	200	368	165	44.83	104	35	33.65	

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Age in years	Total no. of samples	Female		Male		Total no. of culture positives		
-		Number	Positive	Number	Positive			
12-20	55 (11.65%)	51	18	4	2	20 (10%)		
21-40	239 (50.64%)	180	96	59	22	118 (59%)		
41-60	125 (26.48%)	92	36	33	9	45 (22.5%)		
61-80	49 (10.38%)	42	15	7	2	17 (8.5%)		
>80	4 (0.85%)	3	0	1	0	0 (0%)		
Total	472 (100%)	368	165	104	35	200 (100%)		

Table 2 : Age wise distribution of samples and their positivity.

Maximum number of samples (50.64%) were from the age group 21-40 years and maximum positivity (59%) was from the same age group. (Table 2)

Oraganisms	Number	Percentage (%)
Gram negative bacilli	149	74.5
Escherichia coli	64	32
Klebsiella species	61	30.5
Proteus species	07	3.5
Citrobacter species	4	2
Acinetobacter species	2	1
Pseudomonas aeruginosa	11	5.5
Gram positive bacteria	42	21
Staphylococcus aureus	26	13
Coagulase negative staphylococci (CoNS)	11	5.5
Streptococcus pyogenes	1	0.5
Enterococci species	4	2
Fungus	9	4.5
Candida albicans	9	4.5
Total	200	

**Table 3:** Microbial profile of UTI

Table 3 shows that , the isolates included both gram positive and gram negative organisms. Of the 200 isolates, gram negative bacteria accounts for 149(74.5%) while gram positive bacteria accounts for 42(21%).

Bacteria	Antibiotics									
	А	Ak	Caz	Ci	Cot	Gen	Nor	Ni	PIT	Ι
Escherichi coli (n=64)	10	52	45	31	30	45	39	48	41	50
Klebsiella spp (n=61)	11	50	37	31	28	40	37	49	40	49
Proteus spp (n=7)	2	6	5	4	2	4		5	5	6
Citrobacter spp (n=4)	1	4	2	2	2	3	3	3	3	3
Acinetobacte spp (n=2)	0	2			1	1	2	2	2	2
Ps.aeruginosa (n=11)	3	9	7	4	3	5	9	9	9	10
Staph.aureus (n=26)	5	21		18	11	15	20	10		
CoNS (n=11)	3	8		7	5	4	6	9		
Strept. Pyogenes (n=1)	1			1	1		1	1		
Enterococci (n=4)	2	3				2	4	4		

 Table 4 : Antibiotic Susceptibility pattern of bacterial isolates from urine samples of patients

A – Ampicillin, Amc – Amoxyclav, Ak- Amikacin, Caz- Ceftazidime, Ci- Ciprofloxacin, Cot- Co-trimoxazole, G- Gentamicin, Ni-Nitrofurantoin, Nor- Norfloxacin, PIT- Piperacillin+Tazobactem , I- Imipenem, Streptococcus

The most effective antibiotic in case of gram negative bacteria was Amikacin followed by Nitrofurantoin, Piperacillin+Tazobactem and Gentamicin while in case of gram positive bacteria most effective antibiotic was Amikacin followed by Norfloxacin and ciprofloxacin. (Table 4) Antifungal susceptibility testing of *Candida* spp. isolates was not done.

#### III. Discussion

UTI accounts for huge burden on health care systems due to high prevalence of infection in both community and nosocomial settings. UTI is caused by variety of pathogens including *E. coli, Klebsiella spp, Proteus spp, Staphylococcus aureus, Coagulase negative staphylococci* including *Staphylococcus saprophyticus* and *Staphylococcus epidermidis* and also *Candida spp.* Continuous surveillance of antibiotic susceptibility patterns of uropathogens at local level is crucial in dealing with emerging problems of antibiotic resistance and provides assistance in managing effective initial therapy.<sup>1</sup>

In present study urine culture positivity was 42.37% which is very similar to study done by Anup Shah et al  $2015(43.07\%)^{12}$ , Shaista Banoet al 2014 (41.18%)<sup>1</sup> and Telkar et al 2015 (40%).<sup>14</sup> Some studies reported higher positivity like V. Vijaya Swetha et al 2014 (61.91%), Razak S.K et al (2012) (71.72%) and Das RN et al (2006) (71.6%).<sup>14,15,16</sup>

The age and sex wise analysis showed a higher incidence of urinary tract infection in 21-40 years age group in females which can be explained by the fact that urinary tract infections are more common in the reproductive age group. This is because in females the urethra has close proximity and the colonic bacteria have an easy access to it. Females are more prone to UTI than males as they lack bacteriostatic property of prostatic secretions.<sup>17,18</sup>

In our study also most of the samples (50.64%) were from the age group 21-40 years and maximum positivity (59%) was from the same age group. This correlates with other studies by Bashir MF et al (2008)<sup>19</sup> and Getenet B. et al (2011).<sup>20</sup> In a studies conducted by Beyene G et al (2011) and V. Vijaya Swetha et al 2014 reported 53.5% and 48.34% were in the age group between 21-40 years respectively.<sup>21,14</sup>

Like the study by Lekha Tuli et al, the maximum patients presented with the complaint of burning micturition, fever, and abdominal discomfort.<sup>22</sup>

In present study gram negative bacilli 149 (74.5%) were the predominant bacterial isolates followed by Gram positive cocci 42 (21%) which is consistent with another studies. <sup>23,24,25</sup> Overall the most common bacterial isolates were *Escherichia coli* 64(32%) followed by *Klebsiella* species 61(30.5%), *Staphylococcus aureus* 26(13%), Coagulase negative staphylococci (CoNS) 11(5.5), *Pseudomonas aeruginosa* 11(5.5%) *Proteus species* 7(3.5%), *Citrobacter species* 4(2%), *Enterococci species* 4(2%), *Acinetobacter species* 2(1%), *Streptococcus pyogenes* 1(0.5%).

Like our study *Klebsiella spp* was reported as the second most frequently implicated organism after *E.coli* in UTI in some other studies.<sup>26,27</sup> Also *E.coli* (41.73%) was the most common isolated organism in study conducted by Shwetha et al.<sup>14</sup> This was in correlation with other studies by Gupta K.D et al (1999), Moges et al (2002) and Sibi et al (2011).<sup>28,29,4</sup> And the second most common isolated pathogen was *Klebsiella spp* in study conducted by Shwetha et al accounting for 22.90%. This correlates with the studies of Khameneh et al (2009) and Chin et al (2011).<sup>14,30,31</sup>

But Ehinmidu (Ehinmidu, 2003) and Aboderin et al. (2009) recorded *P. aeruginosa* and *Klebsiella spp*, respectively as the predominant bacteria.<sup>26,32</sup> *P. aeruginosa* was also reported as the second most common bacterial isolate in UTI from studies in India by Tambekar et al. (2006)<sup>33</sup> and Lafia, Nigeria by Kolawole et al. (2009).<sup>34</sup>

In our study we found 4.5% isolates were *Candida spp* while Venktesh et al noted 6% isolates were *Cadida spp*.<sup>35</sup>

In this study, among gram negative bacilli isolated from UTI, *E.coli* was sensitive to imipenem, nitrofurantoin, amikacin and piperacillin-tazobactum. A high level of bacterial resistance was seen to ampicillin , ciprofloxacin and ceftazidime. Studies conducted by Mangaiarkkarasi et al. (2013) and Shalini et al. (2011) showed similar antibiotic pattern.<sup>36,37</sup> Amikacin, imipenem, piperacillin-tazobactum, norfloxacin, and nitrofurantoin were more effective against Pseudomonas spp. and Acinetobacter spp. isolated in present study.

Nitrofurantoin is a urinary antiseptic. In their study Joshi et al. (2011) discussed very positive outcomes with Nitrofurantoin as it is cost effective and easily available in developing countries. Besides it can be used safely in pregnancy<sup>38</sup> and causes narrow tissue distribution.<sup>39</sup> In their study, Shaifali et al. (2012) from Lucknow also found the isolates exhibiting maximum susceptibility to Nitrofurantoin.<sup>40</sup>

The high prevalence of resistance to the community used antibiotics such as ampicillin, ciprofloxacin and tetracycline was also noted in some studies.<sup>41,42,43</sup>

Niranjan et al. (2014) reported multiple drug resistance in uropathogenic *E.coli*, which could be due to its widespread prevalence in the community.<sup>44</sup> Lack of guidelines for appropriate use of Antimicrobial agents is another factor responsible for spread of resistance.<sup>45</sup>

In this study, the frequency of ESBL producing organisms among gram negative bacterial isolates was found to be 30 %. A similar frequency of ESBL producing organismswas observed by Telkar et al (29.03%), Dugal et al 2013 (27.67%) and Garg et al (25%).<sup>13,46,24</sup> Detection of ESBL isolates is essential as these ESBL producing gram negative bacilli appear susceptible in-vitro to certain  $\beta$ Lactam antimicrobial agents, yet results in treatment failures. So, proper identification is necessary.<sup>13</sup>

In our study gram positive cocci were sensitive to amikacin followed by norfloxacin, nitrofurantoin and ciprofloxacin.

The prevalence of MRSA in urinary isolates in our study was 28%. While other studies from India found 31.1% (Rajaduraipandhi et al), 54.85% (Anupurba et al) and 66.6% (Garg et al) MRSA strains in urine samples.<sup>47,48,24</sup>

The present study gives an idea about the common trend of increasing antibiotic resistance of uropathogens in this region which could be due to indiscriminate or under dose of antibiotic use. Thus this data may help the physician in proper treatment of urinary tract infections and avoid use of resistant antibiotics.

#### IV. Conclusion

The pattern of antimicrobial resistance of bacteria producing UTI varies in different regions. Monitoring of antimicrobial susceptibility can aid clinicians for prescribing appropriate antibiotics and in prevention of development of drug resistance.

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