

Original Research Article

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Bacteriological Profile and Antibiogram of Uropathogens from a Tertiary Care Hospital: A Two Year Retrospective Analysis

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ABSTRACT

Urinary tract infections (UTIs) are one of the most common bacterial infections encountered in clinical practice. The knowledge of etiology and antibiogram of uropathogens helps in starting empirical treatment till the results of antibiotic susceptibility is awaited. Therefore this two year retrospective study was undertaken to determine the bacteriology and antimicrobial susceptibility pattern of UTIs in a tertiary care hospital. Urine samples from patients suspected of having UTI were processed according to standard microbiological techniques. Bacterial pathogens were isolated, identified and antimicrobial susceptibility testing was done by Kirby Bauer Disc Diffusion from 286 culture positive samples. *Escherichia coli* with 186(65%) isolates was predominant followed by *Klebsiella pneumoniae* 30(10.5%), *Pseudomonas aeruginosa* 20(7%), *Enterococcus* spp. 13(4.6%), *Proteus* spp. 10(3.5%), *Acinetobacter* spp. 8(2.8%), *Staphylococcus aureus* 7(2.4%) and Coagulase negative Staphylococci (CoNS) 6(2.1%). Antibiogram of these bacteria suggests that empirical therapy to cover gram negative bacteria can be started with imipenem or piperacillin /tazobactam or nitrofurantoin. In selected cases vancomycin or linezolid can be added to give gram positive coverage. But in view of the increasing drug resistance antimicrobial susceptibility should be done and definitive therapy started immediately.

Keywords

UTI, Uropathogen,
Bacteriology,
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Introduction

Urinary tract infections (UTI) include various conditions ranging from asymptomatic bacteriuria to severe renal infections resulting into sepsis and can affect any part of the urinary tract from the bladder to the kidney (Kumar *et al.*, 2016, and Flores-Mireles *et al.*, 2015). It is estimated that annually, worldwide 7 million and 1million patients with UTI

attend the outpatient and emergency department respectively. Whereas 100,000 hospitalizations occur annually due to UTI (Wilson *et al.*, 2004). Most of the times these UTIs are treated empirically without any antibiotic susceptibility testing which leads to increased drug resistance in bacteria against commonly used antibiotics (Chiu, 2013). Also in the last two decades it has been seen that the trend of bacterial isolates obtained and their antibiotic sensitivity pattern keeps on

changing (Ram *et al.*, 2000).

Therefore updated information regarding profile of uropathogens and their susceptibility to various antibiotics should be readily available to the clinicians for the development of local data to start appropriate empirical therapy (Majumder *et al.*, 2018; Oli *et al.*, 2017; Vakilwala *et al.*, 2012 and Ko *et al.*, 2008). So with this background, the present study aimed at investigating the bacterial agents responsible for UTIs in a tertiary care hospital setting and study their antibiotic susceptibility pattern.

Materials and Methods

This retrospective study was conducted in a tertiary care hospital over a period of two years, from January 2016 to December 2017 after approval from institutional ethical committee. Clean catch midstream urine or catheterized urine samples were collected in a wide mouthed sterile screw capped containers from suspected patients of UTI. Urine samples were taken to bacteriology lab and processed immediately within half an hour of collection. Samples were inoculated semiquantitatively with calibrated loop on Blood agar and McConkey agar and incubated aerobically overnight at 37°C. Pure growth of a single microorganism with a colony count of >10⁵ colony forming units (cfu)/mL of urine was considered as significant bacteriuria (Stamm *et al.*, 1983). Further, the significant isolates were identified by conventional biochemical methods according to standard microbiological techniques (Collee *et al.*, 2006). Antibiotic Susceptibility Testing was done on Mueller- Hinton agar by Kirby Bauer's disc diffusion method, according to the CLSI guidelines, 2016. The following antimicrobial discs (µg) were used: amoxicillin clavulanic acid (20/10µg), piperacillin tazobactam (100/10µg), amikacin (30µg), co-trimoxazole (25µg), gentamicin

(10µg), tobramycin (10µg), nitrofurantoin (300µg), cefepime (30µg), cefotaxime (30µg), cefuroxime (30µg), ceftazidime (30µg), imipenem (10 µg), Polymyxin B(300U), Penicillin(10U), ciprofloxacin (5µg), norfloxacin (10µg), vancomycin (30µg), Linezolid (15µg). *Escherichia coli* ATCC 25922 *Staphylococcus aureus* ATCC 25923 were used as control strains.

Data management and statistical analysis

Statistical Package for Social Science (SPSS) Software, version 16 was used for data analysis. Chi-square test was performed to obtain the correlations between variables.

Results and Discussion

During the study period, 984 urine samples were analyzed out of which 286 (29.1%) were culture positive. Of these culture positive samples, 173 (60.5%) were from female patients and 113 (39.5%) from male patients. Of the 286 isolates, 254 (88.8%) were Gram negative bacilli, 26 (11.2%) were Gram positive cocci and 6 (2.1%) were *Candida spp.* Amongst the Gram negative isolates, *Escherichia coli* with 186(65%) was predominant followed by *Klebsiella pneumoniae* 30 (10.5%), *Pseudomonas aeruginosa* 20 (7%), *Proteus spp.* 10(3.5%) and *Acinetobacter spp.* 8 (2.8%), (p=0.0001 significant). Amongst Gram positive isolates, *Enterococcus spp.* 13(4.6%) was commonest followed by *Staphylococcus aureus* 7(2.4%) and Coagulase negative *Staphylococcus* 6(2.1%) (Fig. 1).

As far as antibiogram of gram negative bacteria is concerned, Imipenem was the most effective drug with sensitivity ranging from 80-100%. Piperacillin/tazobactam showed good sensitivity against *Proteus spp* (80%) and *E.coli* (77.4%). Amikacin had good sensitivity profile against *E. coli* (79.6%) and

Proteus spp. (70%). Nitrofurantoin was also an effective antibiotic against *E. coli* with 81.2% sensitivity. Polymyxin B was quite effective against *Pseudomonas* spp. and *Acinetobacter* spp with 95% and 87.5% sensitivity respectively. Gram negative bacterial isolates were highly resistant to cotrimoxazole, fluoroquinolones and cephalosporins like cefepime, cefotaxime, ceftazidime, cefuroxime (Table 1).

Gram positive isolates showed 100% sensitivity to Linezolid and Vancomycin. Nitrofurantoin also turned out to be effective with *Staphylococcus aureus* showing 100% sensitivity and CoNS 83.3%. Fifty or less than fifty percent gram positive isolates were resistant to Norfloxacin. Majority of the gram positive bacteria were resistant to Penicillin and Co-trimoxazole (Table 2).

This study gives an insight into UTI, one of the most common infections leading to an antibiotic prescription from a tertiary care hospital. The culture positivity rate was 29.1% from 984 urine samples received from the patients attending the hospital with the symptoms suggestive of UTI. This prevalence rate was similar to various national and other studies from the developing world (Majumder *et al.*, 2018; Khadka *et al.*, 2012; Joshi *et al.*, 2016; Thattil *et al.*, 2018 and Mandal *et al.*, 2012). Greater prevalence of UTI was seen in females accounting for 60.5% of the positive samples, similar to other studies worldwide (Khadka *et al.*, 2012; Joshi *et al.*, 2016; Razak *et al.*, 2012; Singh *et al.*, 2017; John *et al.*, 2015 and Mohammed *et al.*, 2016). The predominant uropathogen in our study was *E. coli* followed by *K. pneumoniae* which is in agreement with many other studies from India and abroad (Majumder *et al.*, 2018; Vakilwala *et al.*, 2012; Khadka *et al.*, 2012; Thattil *et al.*, 2018; Razak *et al.*, 2012; John *et al.*, 2015 and Mohammed *et al.*, 2016).

In the present study imipenem was found to be

effective against more than 80% gram negative isolates as reported by numerous other studies (Majumder *et al.*, 2018; Thattil *et al.*, 2018; Singh *et al.*, 2017; Mohammed *et al.*, 2016; Vecchi *et al.*, 2013 and Rangari *et al.*, 2015). Piperacillin/ tazobactam showed a sensitivity of around 80% for *E.coli* and *Proteus* spp. isolates and around 65% for *Pseudomonas* which is in concordance with many previous studies (Thattil *et al.*, 2018; Vecchi *et al.*, 2013; Rangari *et al.*, 2015 and Singla *et al.*, 2015). This drug was least effective against *Acinetobacter* as reported by Berry *et al.*, (2013). Amikacin also proved to be a good alternative against *E. coli*, *Proteus* spp. and *Pseudomonas* spp. but had limited activity against *Acinetobacter* isolates which is in harmony with other studies (Vakilwala *et al.*, 2012; Joshi *et al.*, 2016; Razak *et al.*, 2012 and Vecchi *et al.*, 2013). Nitrofurantoin turned out is very active against *E. coli* which is the predominant urinary isolate similar to previous reports (Majumder *et al.*, 2018; Rangari *et al.*, 2015 and Singla *et al.*, 2015 and Vecchi *et al.*, 2013). High percentage of resistance against Nitrofurantoin was seen amongst isolates of other gram negative spp. as reported by Joshi *et al.*, (2016) and Thattil *et al.*, (2018). Gentamycin demonstrated a sensitivity rate of 50-55% amongst most of the species similar to few other studies (Joshi *et al.*, 2016; Mandal *et al.*, 2012 and Rangari *et al.*, 2015). Majority of the gram negative isolates were resistant to commonly used cephalosporins and co-trimoxazole as also reported by other researchers (Vakilwala *et al.*, 2012; Razak *et al.*, 2012; Mohammed *et al.*, 2016; Rangari *et al.*, 2015 and Singla *et al.*, 2015 and Berry *et al.*, 2013). Fluoroquinolones were also ineffective against majority isolates similar to previous reports (Razak *et al.*, 2012; Rangari *et al.*, 2015 and Berry *et al.*, 2013). Amoxycillin/ clavulanate proved to be least effective in treating gram negative UTI which is in concordance with some other studies done by Majumder *et al.*,

(2018) and Thattil *et al.*, (2018). As far as gram positive cocci are concerned vancomycin and linezolid were the most effective with 100% sensitivity as reported from many other places (Thattil *et al.*, 2018; Singla *et al.*, 2015; Vecchi *et al.*, 2013 and Rangari *et al.*, 2015). The next effective drug

against UTI due to gram positive cocci was nitrofurantoin which is similar to previous reports by Singla *et al.*, (2015) and Vecchi *et al.*, (2013). Very high resistance was seen to ciprofloxacin amongst enterococcal isolates which was in line with other studies by Thattil *et al.*, (2018) and Rangari *et al.*, (2015).

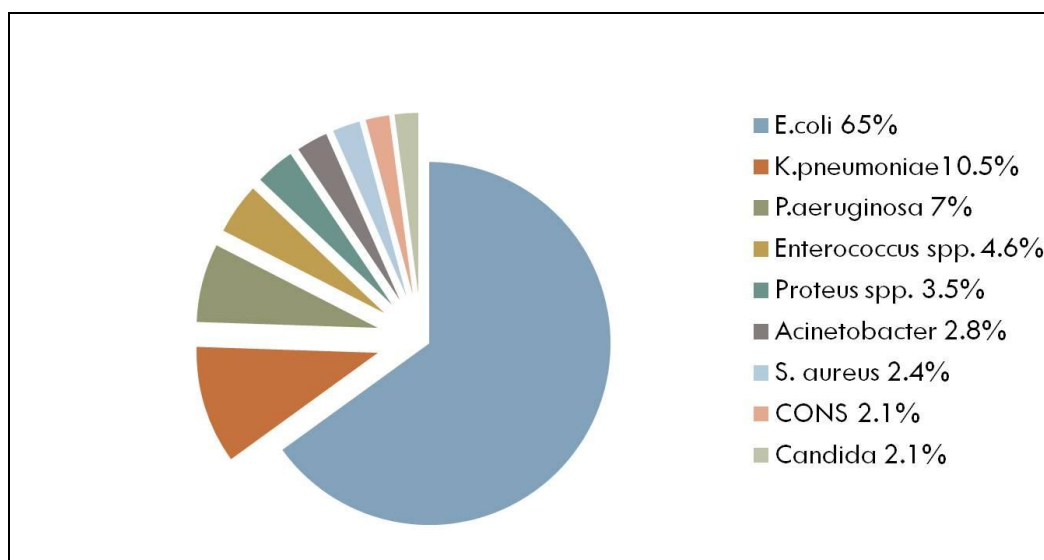
Table.1 Antibiotic sensitivity profile of Gram negative isolates (N=254)

Antimicrobial agent	<i>E.coli</i> (N=186)	<i>K.pneumoniae</i> (N=30)	<i>Pseudomonas</i> (N=20)	<i>Proteus</i> (N=10)	<i>Acinetobacter</i> (N=8)
Amoxicillin/ clavulanate	25(13.4%)	0(0%)	1(5%)	3(30%)	1(12.5%)
Piperacillin/ tazobactam	144(77.4%)	12(40%)	13(65%)	8(80%)	2(25%)
Amikacin	148(79.6%)	10(33.3%)	10(50%)	7(70%)	2(25%)
Co-trimoxazole	48(25.8%)	9(30%)	6(30%)	0(0%)	4(50%)
Gentamycin	105(56.5%)	8(26.7%)	11(55%)	5(50%)	4(50%)
Tobramycin	-	-	10(50%)	5(50%)	6(75%)
Nitrofurantoin	151(81.2%)	2(6.7%)	3(15%)	5(50%)	2(25%)
Ciprofloxacin	56(30.1%)	10(33.3%)	6(30%)	3(30%)	1(12.5%)
Norfloxacin	30(16.1%)	5(16.7%)	5(25%)	2(20%)	2(25%)
Cefepime	49(26.3%)	5(16.7%)	5(25%)	2(20%)	1(12.5%)
Cefotaxime	39(21%)	2(6.7%)	5(25%)	4(40%)	3(37.5%)
Cefuroxime	21(11.3%)	2(6.7%)	1(5%)	2(20%)	1(12.5%)
Ceftazidime	45(24.2%)	3(10%)	6(30%)	2(20%)	2(25%)
Imipenem	185(99.5%)	28(93.3%)	16(80%)	10(100%)	8(100%)
Polymixin B	-	-	19(95%)	-	7(87.5%)

Table.2 Antibiotic sensitivity profile of gram positive isolates (N=26)

Antimicrobial agent	<i>S. aureus</i> (N=7)	CONS (N=6)	<i>Enterococcus spp.</i> (N=13)
Penicillin	1(14.3)	2(33.3)	3(23.1)
Co-trimaxazole	2(28.6)	1(16.7)	5(38.5)
Gentamicin	5(71.4)	4(66.7)	4(30.8)
Ciprofloxacin	3(42.9)	4(66.7)	0(0)
Norfloxacin	2(28.6)	3(50)	4(30.8)
Nitrofurantoin	7(100)	5(83.3)	9(69.2)
Vancomycin	7(100)	6(100)	13(100)
Linezolid	7(100)	6(100)	13(100)

Fig.1 Distribution of culture isolates



Sensitivity of *Staphylococcus aureus* to ciprofloxacin was 42.9% which is comparable to other studies from UP and Nepal (Khadka *et al.*, 2012 and Rangari *et al.*, 2015). Gentamycin turned out to be effective against 70% of the staphylococcal isolates as reported by other workers (Khadka *et al.*, 2012; Vecchi *et al.*, 2013 and Rangari *et al.*, 2015). On the contrary *Enterococcus* isolates showed high resistance to gentamicin which has also been reported in few studies (Joshi *et al.*, 2016 and Thattil *et al.*, 2018). Co-trimoxazole and penicillin were also ineffective against majority of the isolates as in other studies (Khadka *et al.*, 2012; Vecchi *et al.*, 2013 and Thattil *et al.*, 2018).

Therefore this study concludes that *E. coli* is the most likely organism encountered in UTI and most of the strains isolated from a tertiary care hospital are multi drug resistant. Empirical therapy to cover gram negative bacteria can be started with imipenem or piperacillin /tazobactam or nitrofurantoin. In selected cases vancomycin or linezolid can be added to give gram positive coverage. But in view of the increasing drug resistance, antimicrobial susceptibility should be done and definitive therapy started immediately.

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