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A Bacteriological profile and antimicrobial sensitivity pattern in urinary tract infection among reproductive age group of females

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Abstract
Background: The present study was done to isolate and identify of causative agents of urinary tract infection and their antimicrobial susceptibility pattern.
Introduction: Urinary tract infection (UTI) is the second most common infection in human after respiratory tract infection. UTI is more common in females than males. E. coli is most common organism responsible for urinary tract infection.
Materials and Methods: A total 1102 urine samples were collected, out of these samples, 463 urine samples were collected from female patients of reproductive age group (15-45 years). Urine samples were inoculated on MacConkey, CLED (Cysteine Lactose, electrolyte deficient) and Blood agar incubated at 37◦C for 24 hrs, and colonies were processed according to standard procedures
Results: A total 463 urine samples were processed during the study period, from female patients of reproductive age group. 172 (37.1%) had significant bacterial growth, of which 54.65% belonged to 15-25 years of age group followed by 23.25% belonged to 26-35 years and 22.10% belonged to 36-45 years of age group. E. coli are the most common pathogens causing UTI. 44.2% bacterial isolates were identified as E. coli followed by Staphylococcus aureus 18.6%, Klebsiella pneumoniae 11.6%, Coagulase Negative Staphylococci 9.9%, Streptococcus species 5.2%.
Conclusion: Prevalence of UTI was seen higher in Group A (15-25) as compared to Group B (26-35) and Group C (36-45). Both Gram Positive and Gram Negative bacteria are responsible for UTI with dominance of Gram Negative bacteria. In case of Gram Negative bacteria, Cephalosporins showed high resistance while the sensitivity rate of Carbapenems was good. Similarly resistance rate of Penicillin, Ampicillin was high in case of Gram Positive organisms and Vancomycin, Linezolid, Nitrofurantoin showed better sensitivity pattern.

Keywords: urinary tract infection, females, antimicrobial, sensitivity, resistance

Introduction
Urinary tract infection (UTI) is the second most common infection in human after respiratory tract infection [1]. It is defined as the presence of microbial pathogens in the urinary tract with associated symptoms [2]. It is the disease caused by microbial colonization and invasion of the urinary tract that extends from the renal cortex of the kidney to the urethral meatus. UTI is categorized by the site involvements of the infection as; Lower UTI includes; Urethritis (urethra), cystitis (urinary bladder) and Prostatitis (prostate gland) and Upper UTI includes; Acute pyelitis and acute pyelonephritis(kidney) [1]. UTI is more common in females due to presence of short length of urethra and close proximity of urethral meatus to anus. The other predisposing factors being use of contraceptive methods, menopause, pregnancy, urinary tract abnormalities, neurogenic bladder dysfunction, blockage in the tract due to kidney stones or benign prostatic hyperplasia, suppressed immune system, catheterization or any surgical intervention of the urinary tract [3].
E. coli is most common organism responsible for UTI accounting for 70–80% of total cases of UTI. Other organisms such as Staphylococcus spp., Klebsiella spp., Proteus spp, Pseudomonas spp., Enterococcus spp., Enterobacter spp, Acinitobacter spp., Citrobacter spp., Streptococcus spp. etc are the important causative agents of UTI. Candida albicans may also causes UTI [4]. Urinary tract infections may be asymptomatic and symptomatic, both asymptomatic and symptomatic UTIs pose a serious threat to public health care. The symptoms of UTIs such as fever, burning sensations while urinating, LAP, itching, formation of blisters and ulcers in the genital area, genital and suprapubic pain, and pyuria generally depend on the age of the person infected and the location of the urinary tract infected [5]. UTI affects both genders, Females of the reproductive age group (15-45 years) are the most vulnerable, may be due to their anatomical structure of urogenital system, short urethra and close proximity of urethral meatus to anus [6] and certain behavioral factors which include delay in micturition, sexual activity and the use of contraceptives which promote colonization of the periurethral area with coliform bacteria [7]. In this study we will investigate the causative agents of UTI and their antimicrobial susceptibility patterns.

Materials and Methods
Sample Collection and processing
A total 1102 Urine sample were collected from patients attending various outpatients and inpatients department in the Bacteriology laboratory in Department of Microbiology

at National Institute of Medical Sciences & Research, Jaipur, Rajasthan. A total 463 Urine sample were collected from reproductive age group (15-45 years) of females. All Urine samples were processed for isolation and identification of Various microorganisms causing Urinary tract infection (UTI), within the time period of August 2018 to July 2019. All urine samples were collected in wide mouth universal container under sterile condition, Mid-stream urine samples, suprapubic aspiration and catheter tube aspiration were collected and processed.

Culture and identification of isolates

Urine samples were inoculated on MacConkey, CLED (Cysteine Lactose, electrolyte deficient) and Blood agar incubated at 37°C for 24 hrs, and colonies were processed according to standard procedures. The organisms were identified on the basis of colony morphology, and colonies counted for determination of significant or insignificant bacteriuria. A growth of $\geq 10^{5}$ colony forming units/ml was considered as significant bacteriuria, suggestive of UTI [8] Identification was done based on Gram staining, biochemical reactions and other laboratory tests [9].

Antimicrobial susceptibility testing

Antimicrobial susceptibility testing was done by Modified Kirby Bauer Disc Diffusion method on Muller Hinton Agar as per the CLSI guidelines (2018) [10]. The antibiotics tested were Amikacin (AK), Piperacillin + Tazobactam (PIT), Gentamicin (GEN), Tetracycline (TE), Meropenem (MRP), Imipenem (IPM), Ceftazidime (CAZ), Cefotaxime (CTX), Ceftriaxone (CTR), Ciprofloxacin (CIP), Aztreonam (AT), Norfloxacin(NX), Nitrofurantoin(NIT), Penicillin-G (P), Ampicillin (AMP), and Vancomycin (VA), Linezolid (LZ), Erythromycin(E), Clindamicin(CD), Cefoxitin (CX). Results were measured and recorded as compared to that of the manufacturer interpretation charts according to the Clinical and Laboratory Standard Institute (CLSI) guideline (2018) [10].

Result

During the study period, a total 1102 urine samples were collected in the Bacteriology laboratory in Department of Microbiology, National Institute of Medical Sciences & Research, Jaipur. Out of these samples, 463 urine samples were collected from females patients of reproductive age group (15-45 years). Among these 463 urine samples, 283 urine samples were sterile on culture. 180 urine samples showed positive culture growth. Among the 180 isolated strains from culture, 172 isolates were identified as bacterial isolates and 8 isolates were identified as fungal isolates, which were excluded from the study. Reproductive age (15-45) was divided into 3 groups: Group A (15-25), Group B (26-35), Group C (36-45). Out of 172 isolates, 94 bacterial strains were isolated from Group A, followed by 40 bacterial strains from Group B and 38 bacterial strains from Group C. Out of the 172 bacterial strains, 114 isolates were Gram Negative isolates while 58 were Gram Positive isolates. 76 (44.2%) bacterial isolates were identified as E. coli followed by 32 (18.6%) isolates identified as staphylococcus aureus, 20 (11.6%) isolates identified as Klebsiella pneumoniae, 17 (9.9%) isolates were identified as Coagulase Negative Staphylococci, 9 (5.2%) isolates were identified as Streptococcus species, 7 (4.1%) isolates were identified as Citrobacter species, 6 (3.48%) isolates were identified as Pseudomonas species and 5 (2.90%) isolates were identified as Proteus species. E. coli was the main organism causing UTI in all 3 age groups followed by Staphylococcus aureus, Klebsiella pneumoniae and Coagulase Negative Staphylococci.

In the Antimicrobial Susceptibility Pattern of Gram Negative Bacteria, the maximum sensitivity was shown with Meropenem 93.85% followed by Piperacillin+tazobactam 85.1%, Amikacin 80.7%, Imepenem 79.8%, Nitrofurantoin78.9%, Tetracycline 73.7%, Gentamicin 67.5%, Norfloxacin 54.4%, Aztreonam 45.6%, Ceftazidime 44.7%, Cefotaxime 43.0%, Ceftriaxone 40.3%, minimum sensitivity shown by Ciprofloxacin 37.7%.

In the Antimicrobial Susceptibility Pattern of Gram Positive Bacteria, the maximum sensitivity was shown with Vancomycin and Linezolid 100% each followed by Nitrofurantoin87.9 %, Gentamicin 70.7 %, Amikacin, Clindamicin and Cefoxitin 62.7 % each, Ampicillin, Erythromycin and Ciprofloxacin 55.2 % each, Norfloxacin 46.5 %, minimum sensitivity shown by Penicillin –G 44.2 %.

Table 1: Division of all organisms isolated from Urine samples

Total Samples	Reproductive age group samples	Positive Urine Culture	Bacterial Isolates	Fungal Isolates
1102	463	180	172	8

Table 2: Division of bacterial strains according to age groups.

Age Group	Group A (15-25)	Group B (26-35)	Group C (36-45)	Total
Bacterial Strains	94	40	38	172
Percentage	54.65%	23.25%	22.10%	37.1 %

Table 3: Division of Bacterial isolates according to reproductive age groups.

Age Groups / Organisms	Group A (15-25)	Group B (26-35)	Group C (36-45)	Total isolates	Percentage %
E. coli	45	15	16	76	44.2
S. aureus	22	6	4	32	18.6
K. pneumoniae	8	4	8	20	11.6
CONS	9	4	4	17	9.9
Streptococcus spp.	2	5	2	9	5.2
Citrobacter spp.	3	3	1	7	4.1
Pseudomonas spp.	3	1	2	6	3.5
Proteus spp.	2	2	1	5	2.9
Total	94	40	38	172	100

Table 4: Antimicrobial Susceptibility Pattern of all Bacterial Isolates.

AMA	Gram Negative Isolates (114)		AMA	Gram Positive Isolates (58)	
	S	S%		S	S%
AK	88	77.2	AK	36	62.1
PIT	97	85.1	GEN	41	70.7
GEN	77	67.5	P	32	55.2
TE	51	44.7	AMP	32	55.2

MRP	107	93.8	VA	58	100.0
IPM	91	79.8	LZ	58	100.0
CAZ	51	44.7	E	32	55.2
CTX	49	43.0	CD	36	62.1
CTR	46	40.3	NX	27	46.5
CIP	43	37.7	NIT	51	87.9
AT	52	45.6	CIP	32	55.2
NX	62	54.4	CX	36	62.1
NIT	90	78.9			

Table 5: Antimicrobial Susceptibility Pattern of Gram Negative Bacteria.

AMA	E. coli		K. pneumoniae		Citrobacter spp.		Proteus spp.		Pseudomonas spp.	
	S	S%	S	S%	S	S%	S	S%	S	S%
AK	58	76.3	15	75.0	6	85.7	4	80.0	5	83.3
PIT	65	85.5	17	85.0	6	85.7	4	80.0	5	83.3
GEN	50	65.7	14	70.0	5	71.4	4	80.0	4	66.6
TE	36	47.4	7	46.7	3	42.5	2	40.0	3	50.0
MRP	71	93.4	18	90.0	7	100.0	5	100.0	6	100.0
IPM	60	78.9	15	75.0	6	85.7	5	100.0	5	83.3
CAZ	34	44.7	8	40.0	3	42.8	3	60.0	3	50.0
CTX	35	43.4	7	35.0	3	42.8	3	60.0	3	50.0
CTR	34	44.7	6	30.0	2	28.5	2	40.0	2	33.3
CIP	26	34.2	7	35.0	3	42.8	3	60.0	4	66.6
AT	36	47.3	8	40.0	3	42.8	3	60.0	2	33.3
NX	33	43.4	9	45.0	4	57.1	3	60.0	2	33.3
NIT	67	88.1	14	70.0	5	71.4	4	80.0	N.A.	N.A.

Table 6: Antimicrobial Susceptibility Pattern of Gram Positive Bacteria.

AMA	S. aureus		CONS		Streptococcus spp.	
	S	S%	S	S%	S	S%
AK	21	62.5	10	58.8	5	55.5
GEN	23	71.8	12	70.5	6	66.6
P	15	46.8	8	47.0	9	100.0
AMP	18	56.2	9	52.9	5	55.5
VA	32	100.0	17	100.0	9	100.0
LZ	32	100.0	17	100.0	9	100.0
E	19	59.3	9	52.9	4	44.4
CD	21	62.5	11	64.7	4	44.4
NX	15	46.8	8	47.0	4	44.4
NIT	28	87.5	15	88.2	8	88.8
CIP	17	53.1	10	58.8	5	55.5
CX	19	59.3	13	76.4	4	44.4

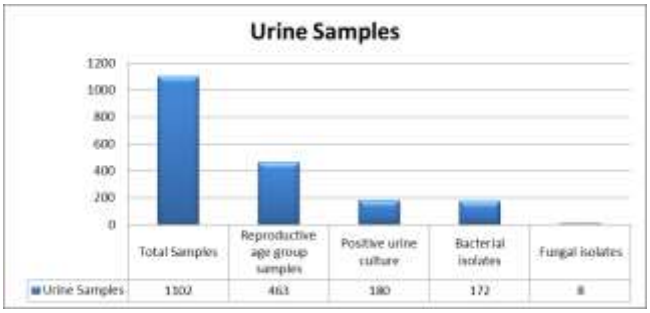

Fig 1: Division of all organisms isolated from Urine samples.

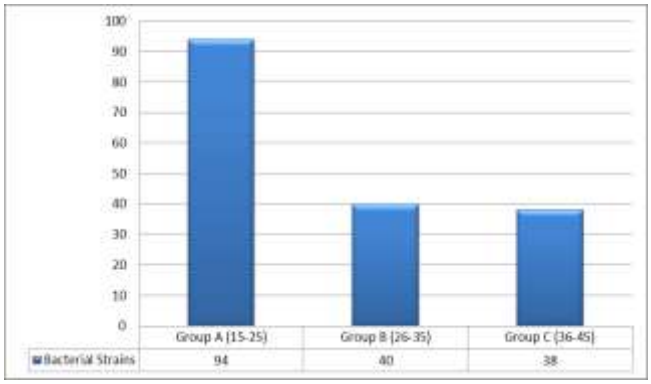

Fig 2: Division of bacterial strains according to age groups.

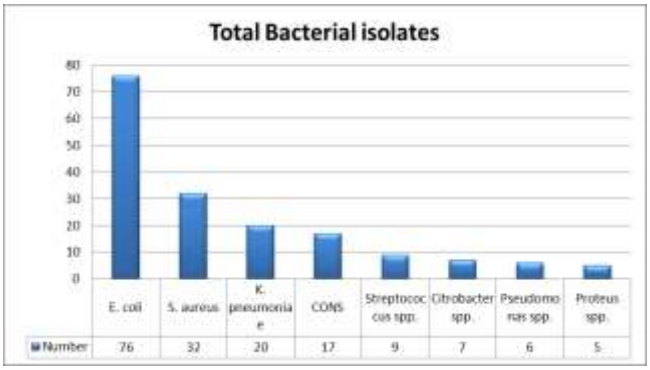

Fig 3: Bacterial strains isolated from urine samples.

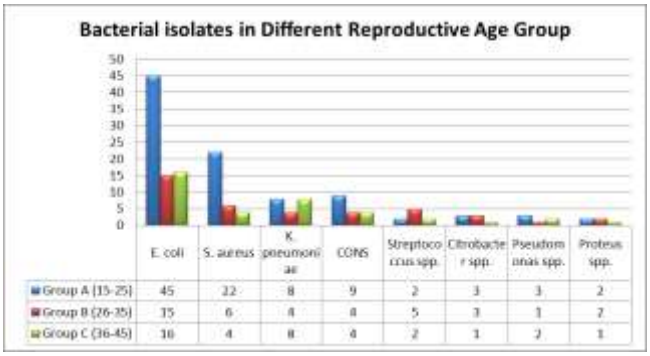

Fig 4: Division of Bacterial isolates according to reproductive age groups.

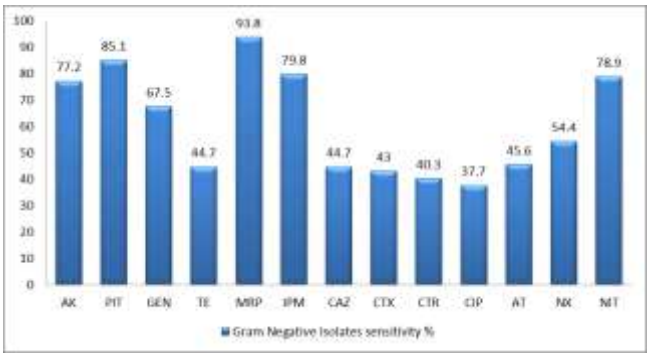

Fig 5: Antimicrobial Susceptibility Pattern of Gram Negative Bacteria.

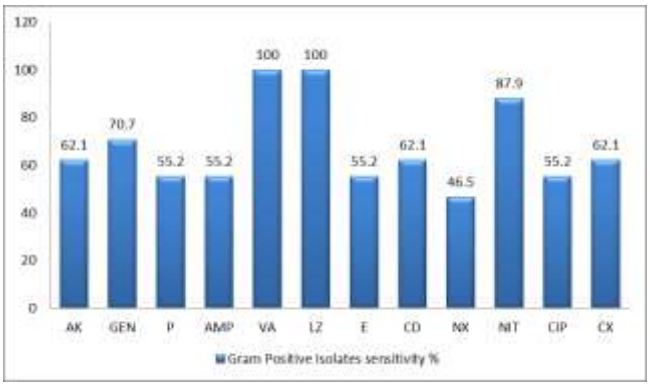

Fig 6: Antimicrobial susceptibility pattern of gram positive bacteria.

Discussion

The present study was carried out in the Department of microbiology, National Institute of Medical Sciences & Research, NIMS University, Jaipur Rajasthan, from August 2018 to July 2019. During this study period, a total 1102 urine samples were collected at NIMS hospital. A total of 463 urine samples were collected from female patients of reproductive age group (15-45 years). Among these 463 urine samples, 283 urine samples were sterile on culture. 180 urine samples showed positive culture growth. Among the 180 isolated strains from culture, 172 isolates were identified as bacterial isolates and 8 isolates were identified as fungal isolates, which were excluded from the study. In this study the prevalence of Urinary tract infections in reproductive age group of females was about 37.1 %, which is quite similar to Martin Odoki *et al*. [11] study i.e. 37.5 %, Chandra Bala Sekhara *et al*. [12] i.e. 31.6 % and Farhat Tahira *et al*. [13] i.e. 49.62%. In present study out of 172 isolates, the maximum bacterial strains isolated from 15-25 years age group of females i.e. 54.65% followed by 23.25% in 26-35 years age group and 22.10 % in 36-45 years age group of females. Similar observation was seen in their study by Muthulakshmi M. *et al*, [14] around 44.0 % in 15-24 years age group of females followed by 36% in 35-44 years age group, and around 20% in 25-34 years age group of females. According to our study E. coli was main causative organism of UTI infections with isolation rate of 44.2 % followed by S. aureus 18.6 %, Klebsiella pneumoniae 11.6 % and Proteus spp. 2.9 %. Which correlates with Martin Odoki *et al*. [11] study, E. coli 40.9 %, Klebsilella pneumoniae 10.6 % and Proteus spp 4.5 % and correlates with Nora Refat Mohamed *et al*. [15] i.e. E. coli 44.2 %, S. aureus 22.1 % and Klebsiella pneumoniae 9.5 %, Proteus spp. 6.3 %, in other study done by Vijay Prakash Singh *et al*[16] E. coli 33.3 %, S. aureus 20.0 % and Klebsiella pneumoniae 13.3 %. Among these isolates, Gram Negative bacteria showed a high susceptibility to Meropenem followed by Pipracillin + Tazobactam, Imipenem, Nitrofurantoin, Amikacin Tetracycline, Gentamicin. and highest resistance rate was seen towards Ciprofloxacin, third generation cephalosporins, Azetreonem and Norfloxacin. This sensitivity pattern correlates to Deepika Atray *et al*. [17] and D. Maheswary *et al*. [18] and Kothari A *et al*. [19] findings. Among these isolate Gram positive bacteria showed a high susceptibility to Vancomycin and Linezolid, Nitrofurantoin, Gentamicin, Amikacin which correlates with study of Vijay Prakash Singh *et al*, [20] Ruhi Khan *et al*. [21] and Sonali Waske *et al*. [22] In our study prevalence of UTI in reproductive age group of females was 37.1 % and other studies have also shown quite similar results, we have divided into different age group of females which suggest that the percentage of Urinary tract infections are more common in 15-25 years age group of females, which may be due to not maintaining proper personal hygiene and lack of general awareness. In our study the resistant rate of Cephalosporins was higher as compared to other studies which indicates the production of Beta lactamases mainly Extended spectrum of beta lactamase.

Conclusions

According to this study, prevalence of UTI was seen higher in Group A (15-25) as compared to Group B (26-35) and Group C (36-45). Both Gram Positive and Gram Negative bacteria are responsible for UTI with dominance of Gram Negative bacteria. Escherichia coli was the main causative organism of UTI and was followed by Staphylococcus aureus in our study. Most of the first line drugs or routinely used drugs were resistant whereas higher drugs were sensitive. In case of Gram Negative bacteria, Cephalosporins showed high resistance while the sensitivity rate of Carbapenems was good. Similarly resistance rate of Penicillin, Ampicillin was high in case of Gram Positive organisms and Vancomycin, Linezolid, Nitrofurantoin showed better sensitivity pattern. So this study will be helpful for clinician to initiate better empirical therapy for such patients and thus it will be helpful in reducing the morbidity rate**.**

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