How to Cite:
Bajpai, S., Tiwari, M. K., Khatri, H. B., & Bhatiani, A. (2021). Clinical and microbiological profile of urinary tract infection in children less than twelve years of the age and their antibiotics resistant pattern at a Tertiary Care Hospital in Kanpur, India. *International Journal of Health Sciences*, *5*(S1), 541–551. https://doi.org/10.53730/ijhs.v5nS1.5926

Clinical and microbiological profile of urinary tract infection in children less than twelve years of the age and their antibiotics resistant pattern at a Tertiary Care Hospital in Kanpur, India

Shweta Bajpai
Tutor, Department of Microbiology, GMC, Kannauj, UP, India
Email: bajpai.shweta.97@gmail.com
ORCID-0000-0002-8912-1911

Manish Kumar Tiwari
Tutor Department of Microbiology, SMC, Unnao, UP, India
Email: mprabhat111@gmail.com

Dr. Himanshu Bhim Khatri
Associate Prof. Department of Microbiology, Surabhi Institute of Medical Sciences, Siddipet, Telangana, India
Email: himanshubkhatri@yahoo.co.in

Dr. Ambika Bhatiani
Professor Department of Microbiology, Rama Medical College, Hospital & Research Centre Kanpur, UP India
Email: ambikabhatianimicrobiology@gmail.com

***Abstract*---**Background: Urinary tract infection (UTI) is the most common bacterial infection which is seen in childhood population, which starts from the calyces of the kidney to the urethra. It is an important cause of morbidity and mortality in children mostly in the first 2 years of life. The Gram negative enteric bacilli, especially Escherichia coli and Klebsiella spp. the dominant pathogens. The objective of the study was to analyze the causative microorganism and their Antibiotic Resistance Pattern in UTI in childhood population between 0-12years of the age. Methods: This one year prospective and observational study was conducted in the Department of Microbiology; Rama Medical College Hospital & Research Centre Kanpur, Uttar Pradesh, India, on 100 pediatric patients (aged 0-12 years). All the pediatric age group (0-12 years) with suspected UTI and, whose urine

International Journal of Health Sciences E-ISSN 2550-696X © 2021.
Corresponding author*: *Bhatiani*, *A.*; *Email:* *ambikabhatianimicrobiology@gmail.com
Manuscript submitted: 27 April 2021, Manuscript revised: 18 June 2021, Accepted for publication: 09 July 2021

culture showed one or two pathogen with colony counts greater than or equal to 105 colony forming units (CFU)/ml was included for the study. Clinical data was obtained from OPD and IPD Patients. Antimicrobial susceptibility was done for positive urine culture by Kirby-Bauer disk diffusion method. The data was expressed in terms of frequency and percentage. Statistics results were analyzed by using Chi-square (x2) test by Graph Pad® Software, Inc. 2236 Avenida de la Playa La Jolla, CA 92037 USA, InStat statistical software. Results: Of the 100 urine samples analyzed, 39% had significant bacteuria. The most predominantly isolated organisms from cases of paediatric urinary tract infections were Escherichia coli 19 (48.75%) followed by Klebsiellaoxytoca 5 (12.82%), Klebsiella pneumoniae 3 (7.69%), Pseudomonas aeruginosa 1 (2.56%), Enterococcus fecalis 5 (12.82%), MSSA 2 (5.12%), C. tropicalis 2 (5.12%) and C. krusei 2 (5.12%). Most of the gram negative strains (68%) showed high resistance towards amoxicillin, Cefotaxime, Ceftriaxone, Gentamycin, Ofloxacin Amikacin, Levofloxacin and least towards nitrofurantoin (21.42%) Norfloxacin, Piperacilin- tazobactum, Imepenem and Meropenem. Enterococcus andMSSAisolates were found 100% susceptible towards Vancomycin, Linezolid, and Teicoplanin. Conclusions: Regular surveillance and monitoring should be done to find out the prevalent isolates and their antibiotic sensitivity pattern to choose empirical antibiotic treatment for urinary tract infections (UTIs).

***Keywords*---**pediatric, Urinary tract infection, Antibiotic resistant, *Escherichia coli.*

Introduction

Urinary tract infection (UTI) is the most common bacterial infection which is seen in childhood population, which starts from the calyces of the kidney to the urethra. It is an important causes of morbidity and mortality in children mostly in the first 2 years of life [1–4].The Gram negative enteric bacilli, especially *Escherichia coli* and *Klebsiella spp.*[5] are the dominant pathogens, followed by *Enterococcus spp.*, *yeasts* and *Staphylococcus aureus* thathave emerged as large group of bacterial agents in recent years [6-8]. The overall incidence of urinary tract infection is reported as 7% in young girls and 2% in boys during the first 6 years of life [9]. The basic objects of treatment of the urinary tract infections in childhood are early relief of symptoms and prevention of complications, like urolithiasis, urosepsis, renal abscess, and permanent damage of renal parenchyma cells [10]. To accomplish these aims, empirical antibiotic therapy is prescribed before the culture results are available [11]. Due to injudicious use of antibiotics worldwide, resistance of pathogens to antibiotics is steadily increasing [12, 13]. These trends of resistance have crucial implications in guiding the empirical antibiotic therapy [14]. The knowledge of the causative agent and their antibiotic resistance patterns in specific geographical regions may assist the clinicians in choosing the appropriate empirical antimicrobial therapy for these infections [15].

Material and Methods

This one year prospective and observational study was conducted in the Department of Microbiology; Rama Medical College Hospital & Research centre Kanpur, Uttar Pradesh, India, on 100 pediatric patients (aged 0-12 years). A suitable statistical analysis was carried out according to the study. The study was conducted accordance with ethical standards which was obtained from the Institutional Ethical Committee of RMCH & RC, and informed consent was obtained from their parents. All the pediatric age group (0-12 years) with suspected UTI and, whose urine culture showed one or two pathogen with colony counts greater than or equal to 10^5 colony forming units (CFU)/ml was included for the study. All pediatric age group who did not satisfy the inclusion criteria was excluded from the study. Mid-stream urine sample was collected with the strict aseptic precautions. Sample was collected before the start of antibiotics, from inpatients (IPD) and out-patients (OPD) suspected to be having urinary tract infection. Samples were processed as soon as received in laboratory. In cases where a delay was expected, the samples were refrigerated for up to 4 hours at 4°C. Urine culture was done by semi-quantitatively methods on CLED agar, 5% sheep Blood Agar and MacConkey Agar plates. The plates were incubated at 37°C for 16-18 hrs. The Standard technique was performed for semi-quantitative culture of urine for colony count. The bacterial isolates were identified based on colony morphology, gram stain and standard biochemical tests. A bacterial suspension was prepared in physiological normal saline solution by picking up 1-2 colonies from culture plates. The bacterial suspension was placed on Mueller-Hinton Agar (MHA) plate by swabbing technique methods. Antibiotic disks were placed onto the cultures medium surface. The culture plates were incubated at 37◦C for 24 hours; Then the zone of inhibition were recorded according to the Clinical and Laboratory Standards Institute (CLSI) guidelines [16].

The following antibiotics were tested for bacterial isolates such as Amoxicillin (20µg), Piperacillin tazobactam(100µg), Cefotaxime(30µg), Ceftriaxone (30µg), Gentamycin (10µg), Amikacin (30µg), Ofloxacin (1µg), Levofloxacin (1µg), Imipenem (10µg), Meropenem(10µg), Colistin(10µg), Nitrofurantoin (50µg), Tigecycline (10µg). for Gram negative organisms and Amoxicillin (20µg) Erythromycin(5µg) Clindamycin (2µg) Tetracycline(10µg), Linezolid (10µg), Vancomycin (5µg), Teicoplanin (30µg), Co-Trimoxazole (25µg) and Nitrofurantoin (50µg). For Gram positive organisms.

Results

A total 100 urine samples were obtained from pediatric patients suspected of having UTI among which 55 were males and 45 were females. In males, most (19%) were found in the age group of (9-12) years and in females, most (11%) were found in the age group of (0-1) (Table-1). The most predominantly isolated organisms from cases of pediatric urinary tract infections were Escherichia coli 19 (48.75%) followed by *Klebsiellaoxytoca* 5 (12.82%), *Klebsiella pneumoniae* 3 (7.69%), *Pseudomonas aeruginosa* 1 (2.56%), *Enterococcus fecalis* 5 (12.82%), MSSA 2 (5.12%), *C. tropicalis* 2 (5.12%) and *C. krusei* 2 (5.12%) (Table-2). According to the age group, most of the isolates were found from the age group 9-12 years; among which E. coli was the dominant organism 11 (57.9%) followed by *Klebsiella*

pneumoniae 2 (66.66%), MSSA 2(100%), E*nterococcus faecalis* 2 (40%), *Klebsiella oxytoca* 1(20%) *and C. tropicalis* 1(50).(Table-3)Maximum numbers of E. coli were isolated from female patients as compared to males and this was statistically when compared to non-E.coli isolates (p <0.000.) (Table-4) Most of the gram negative strains (68%) showed high resistance towards amoxicillin and least towards nitrofurantoin (21.42%). *Enterococcus* and MSSA isolates were found 100% susceptible towards vancomycin, linezolid, and teicoplanin (Table-5).
Comparison among present study and others studies (Table-6).

Table 1
Age and ward wise distribution of clinical samples (n=100)

Age	Total	Male	Female	Ward	
				OPD	IPD
0-1 year	21	10	11	6	15
1-3years	16	10	6	3	13
3-6 years	18	10	8	4	14
6-9 years	16	6	10	3	13
9-12years	29	19	10	10	19
Total	100	55%	45%	26%	74%

Total 100 urine samples were collected from suspected urinary tract infection patients from pediatrics ward of IPD and OPD, Out of which 55 were males and 45 were females. Maximum 19% male patients were found in the age group of 9-12 years and 11% females patients were found in the age group of 0-1 (Table-1).

Table 2
Distribution of Culture Positive Uropathogens (n=39)

Organism	IPD	OPD	Total (n=39)	Percentage
Escherichiacoli	15	04	19	48.75%
Klebsiellapneumoniae	2	01	03	7.69%
Klebsiellaoxytoca	3	02	05	12.82%
Pseudomonasaeruginosa	01	00	01	2.56%
MSSA	02	00	02	5.12%
Enterococcusfaecalis	04	01	05	12.82%
C.tropicalis	02	00	02	5.12%
C.krusei	01	01	02	5.12%

Out of the 39 isolates; Escherichia coli were found dominant isolates 19 (48.75%) followed by *Klebsiellaoxytoca* 5 (12.82%), *Klebsiella pneumonia* 3 (7.69%), *Pseudomonas aeruginosa*1 (2.56%), *Enterococcus fecalis* 5 (12.82%), *MSSA* 2 (5.12%), *C.tropicalis*2 (5.12%) and *C.krusei*2 (5.12%) (Table 2)

Table 3
Age Wise Distribution of Culture Positive Isolate (n=39)

Isolate	Age0-1 Year	Age1-3 Year	Age3-6years	Age 6-9 Years	Age 9-12 Years
Escherichia. Coli	1(5.2%)	–	3(15.8%)	4(21.1%)	11(57.9%)
Klebsiellaoxytoca	–	2(40%)	1(20%)	1(20%)	1(20%)
Klebsiellapneumoniae	–	1(33.33%)	–	–	2(66.66%)
Pseudomonas aeruginosa	–	–	1(100%)	–	–
Enterococcusfaecalis	1(20%)	1(20%)		1(20%)	2(40%)
MSSA	–	–	–	–	2(100%)
C.tropicalis	–	1(50%)	–	–	1(50%)
C.krusei	1(50%)	1(50%)	–	–	–
Total	3(7.7%)	6(15.4%)	5(12.8%)	6(15.4%)	19(48.7%)

Out of 39 isolates, most of the isolates were found from the age group 9-12 yrs; among which E. coli is the dominant organism 11 (57.9%) followed by *Klebsiella pneumonia* 2 (66.66%), MSSA 2(100%), E*nterococcus faecalis* 2(40%), *Klebsiella oxytoca 1(20%) and C. tropicalis 1(50).*(Table-3)

Table 4
Prevalence of E. coli versus Non E. coli bacteria by sex.

Gender	E.coli	Non E.coli	Total	P Value
Male	4	15	19 (48.7%)	X^2=11.35
Female	15	5	20 (51.3%)	P=0.000

Maximum numbers of E. coli were isolated from female patient as compared to male and it is highly significant as compared to non-E.coli; p-value is <0.000.(Table 4.)

Table 5
Antibiotic Resistance Pattern of different bacterial Isolates (n=35)

Antibiotic	MSSA(n-2)%	Enterococcusfaecalis (n-5)%	*Escherichia* coli (N=19)%	Klebsiella Oxytoca (N=5)%	Klebsiellapneumoniae (N=3)%	Pseudomonas aerunginosa (N=1)%
Amoxycilin	1(50%)	3(60%)	13(68.42%)	3(60%)	2(66.67%)	1(100%)
Erythromycin	1(50%)	2(40%)	-	-	-	-
Clindamycin	1(50%)	1(20%)	-	-	-	-
Tetracycline	1(50%)	1(20%)	-	-	-	-
Linezolid	0	0	-	-	-	-
Vancomycin	0	0	-	-	-	-
Teicoplanin	0	0	-	-	-	-
Co-	1(50%)	2(40%)	-	-	-	-

Antibiotic	MSSA(n-2)%	Enterococcusfaecalis (n-5)%	*Escherichia* coli (N=19)%	Klebsiella Oxytoca (N=5)%	Klebsiellapneumoniae (N=3)%	Pseudomonas aerunginosa (N=1)%
Trimoxazole						
Norfloxacin	0	2(40%)	5(26.31%)	1(20%)	1(33.33%)	0
Nitrofurantoin	1(50%)	1(20%)	4(21.1%)	1(20%)	1(33.33%)	0
PiperacilinTazobactum	-	-	6(31.58%)	1(20%)	1(33.33%)	0
Cefotaxime	-	-	8(42.1%)	2(40%)	1(33.33%)	1(100%)
Ceftriaxone	-	-	9(47.37%)	3(60%)	2(66.67%)	1(100%)
Gentamycin	-	-	8(42.1%)	2(40%)	1(33.33%)	0
Amikacin	-	-	9(47.37%)	3(60%)	1(33.33%)	0
Ofloxacin	-	-	7(36.84%)	2(40%)	1(33.33%)	1(100%)
Levofloxacin	-	-	10(52.63%)	2(40%)	1(33.33%)	1(100%)
Imepenem	-	-	5(26.31%)	1(20%)	1(33.33%)	0
Meropenem	-	-	4(21.1%)	2(40%)	1(33.33%)	0

Most of the gram negative strains (68%) showed highly resistant towards amoxicillin, amoxicillin, Cefotaxime, Ceftriaxone, Gentamycin, Ofloxacin Amikacin, Levofloxacin and least towards nitrofurantoin (21.42%) Norfloxacin, Piperacilin Tazobactum, Imepenem and Meropenem. And least towards nitrofurantoin (21.42%), *Enterococcus and MSSA* isolates were found (100%) susceptible towards Vancomycin, Linezolid. and Teicoplanin (TABLE 5).

Table 6
Comparison of Pediatric Urinary Tract Infection from Various Studies

S.No.	Study	Year	Age group	Gender	Isolated Organism
1	Mahmut Abuhandan et al[17]	2013	0-15years	M (13.1%) <F (86.9%)	*E. coli (64.5%), Klebsiella spp. (12.1%), Proteus mirabilis (8.4%), Staphylococcus aureus (4.7%), Pseudomonas aeruginosa (4.7%), Acinetobacter spp. 2.8%) and Enterococcus spp. (2.8%)*
2	Neelam Taneja et al[18]	2010	< 12 years	M (77.8%)> F (22.2%)	*Escherichia coli (47.1%), Klebsiella spp. (15.6%), Enterococcus fecalis*

S.No.	Study	Year	Age group	Gender	Isolated Organism
					(8.7%), members of tribe Proteae (5.9%), Pseudomonas aeruginosa (5.9%) and Candida spp. (5.5%).
3	Chang-Teng Wu et al[19]	2015	0-12 years	M (11.4%)>F (11.2%)	Escherichia coli (68%) Klebsiella pneumoniae (8.1%) and Proteus mirabilis (6.8%)
4	NirmalJitetal [20]	2014	< 1 years	M (60.5%)>F (39.4%	*E.coli (45.4%), Klebsiella (16.7%) and Enterococcus spp (13.2%) candida (21.1%)*
5	Rima h. Hanna-wakim et al [21]	2015	0-18 years	M (22.3%) <F (77.7%)	*E.coli (79.4%) Klebsiella(7.8%) Pseudomonas aeruginosa (2.1%) Proteus spp.(3.8%) Enterococcus (1.5%)*
6	Seyed Reza Mirsoleymani et al[22]	2013	< 16 years	M (54.9%)>F (45.1%)	*E. coli (65.2%), Klebsiella spp. (26%), Pseudomonas aeruginosa (3.6%), and Staphylococcus coagulase positive (3.7%)*
7	Present Study	2017	0-12 years	M(48.7%)<F(51.3%)	*Escherichia coli (48.75%) Klebsiella oxytoca (12.82%), Klebsiella pneumonia (7.69%), Pseudomonas aeruginosa (2.56%), Enterococcus fecalis (12.82%), MSSA (5.12%),C. tropicalis (5.12%) and C. krusei (5.12%)*

Statistical Analysis

The data was expressed in terms of frequency and percentage. Statistics results were analyzed by using Chi-square (x2) test, confidence interval (CI), odds ratio (OR) analysis and *P* value by Graph Pad® Software, Inc. 2236 Avenida de la Playa La Jolla, CA 92037 USA, InStat statistical software. A p-value <0.05 was considered statistically significant, and a p-value <0.01 was considered highly significant.

Discussion

Our study describes the distribution and antibiotic Resistant pattern of uropathogens which is isolated from pediatric population with suspected UTIs in the Rama medical college and hospital Kanpur uttarpradesh. In this study 55% were males and 45% were females. A similar study conducted by Seyed Reza Mirsoleymani, Morteza Salimi, Masoud Shareghi Brojenis, et al [22] male was (54.9%) and female was (45.1%). In our study showed that the prevalence of the rate of urinary tract infections (39%) among pediatric population, which is almost concordant study done by Dash et al., by (34.5%) and (36.6%) by Mehta et al [23,24]. In present study Maximum 19% male patients were found in the age group of (9-12) years and 11% females patients were found in the age group of (0-1). another study which is conducted by GK Rai, HC Upreti, SK Rai, et al [25].The maximum (55.4%) male patients were found in the (11-15) years of the age and (43.5%) female patients were found in the age group of (<1) this varies may be by age, gender, geographical reason and other factors. Gram-negative bacilli were the predominant causative agent for childhood urinary tract infections (UTIs) accounting to 72% in our study. Escherichia coli were more dominant isolates 19 (48.75%) followed by *Klebsiellaoxytoca* 5 (12.82%), *Klebsiella pneumonia* 3 (7.69%), *Pseudomonas aeruginosa* 1 (2.56%), *Enterococcus fecalis* 5 (12.82%), *a nearly similar various study conducted by* Dr Priyanka Saha, Dr Soma Mondalet al[26]Neelam Taneja et al[18] and NirmalJit et al [20] *E. coli (45.13%) was the commonest bacteria which is responsible for urinary tract infections in childhood population.* Antibiotic resistance is a major clinical health care problem for treating infections which is caused by these uropathogens. The resistance to the antimicrobials has increased all over the years. Resistance rates vary from country to country.Antibiotic resistant pattern in our study, most of the gram negative strains (68%) showed highly resistant towards amoxicillin, and least towards nitrofurantoin (21.42%), *Enterococcus and MSSA* isolates were found (100%) *Enterococcus and MSSA* isolates were found (100%) susceptible towards Vancomycin, Linezolid. and Teicoplanin.in the previous study done by Nirmaljit Kaur, Shweta Sharma, Shalini Malhotra,*et al[20] mostly* gram negative isolates (50%) showed high resistance to gentamicin, amikacin, cefotaxime and norfloxacin.

Conclusion

In pediatric urinary tract infections, it is rather dreadful to notable that the almost of the Gram negative strains (68%) were found resistant to one or two antibiotics. Antibiotic resistance is enhancing serious predicament which is menace the lives of hospitalized people and also considerably to the health maintenance cost. For that reason it is a very serious problem to be communicate by the assemblyperson

to formulate accurate antibiotics should be prescribed in our country, and it's also Regular surveillance and monitoring should be done to find out the prevalent isolates and their antibiotic sensitivity pattern to choose empirical antibiotic treatment for urinary tract infections (UTIs) in pediatrics population in our country.

Limitations

The number of participants or observations was very low in our study and the whole spectrum of signs and manifestations were not included in this study.

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