Global Journal of Medical Research: C
Microbiology and Pathology
Volume 14 Issue 7 Version 1.0 Year 2014
Type: Double Blind Peer Reviewed International Research Journal
Publisher: Global Journals Inc. (USA)
Online ISSN: 2249-4618 & Print ISSN: 0975-5888

Microbiological Spectrum and Susceptibility Pattern of Clinical Isolates from Children Suspected of Urinary Tract Infection, Visiting Kanti Children's Hospital, Maharajgung, Kathmandu

By Bidur Aryal, Pappu Kumar Mandal & Prem Dev Tripathi
ST. Xaviers College, Maitighar, Nepal

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Methods: A hospital based cross section study was conducted among children suspected of urinary tract infection in Kanti Children's Hospital over a period of six months from August 2012 to November 2012. A total of 1890both sexes, ranging from post natal period to 14 years of age were studied. The modes of presentation, laboratory investigation reports, which included urine routine microscopy, bacterial isolation with colony count from urine culture, antibiotic sensitivity pattern and multidrug resistant profile, were documented. Data were analyzed by the Chi Square Test.

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GJMR-C Classification : *NLMC Code: QW 4, WJ 151*

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Microbiological Spectrum and Susceptibility Pattern of Clinical Isolates from Children Suspected of Urinary Tract Infection, Visiting Kanti Children's Hospital, Maharajgung, Kathmandu

Bidur Aryal [α], Pappu Kumar Mandal [σ] & Prem Dev Tripathi [ρ]

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Results: Among 1890 urine samples, 300(15.88%) showed culture positive result: 256 gram negative organisms and 44 gram positive organisms. Among the positive growth samples 144(48%) were male patients and 156(52%) were female patients.; *Escherichia coli* was most common organisms isolated (52.33%), followed by *Klebsiella spp* (17.33%), *Staphylococcus aureus*(14%), *Pseudomonas aeruginosa* (4.33%), *Proteus spp* (3.67%), *Citrobacter spp* (3%), *Acinetobacter spp*(1.66%), *Enterobacter spp* and *Hafnia alvei* (1.33%), *Streptococcus spp* (0.66%) and *Salmonella paratyphi* B(0.33%). Nitrofurantoin was found to have the highest sensitivity (71.67%) amongst most bacteria. Amikacin, Norfloxacin and Gentamicin had sensitivity of 69%, 61.71% and 61.67% respectively. *Pseudomonas aeruginosa* was 100% sensitive to Tobramycin, Piperacillin and Imipenem. Though sensitivity to Vancomycin was tested to 44 cases it was 100% sensitive to *Staphylococcus aureus* and *Streptococcus fecalis*. Highest degree of resistance was noted with Ceftazidime (64%), Ofloxacin (61.33%), Ampicillin (60%), ciprofloxacin (55.67%), Cotrimoxazole (52%), Gentamicin (38.33%), Amikacin (28%) and Nitrofurantoin (23.67%).

Conclusion: Infected urine stimulates an immunological and inflammatory response leading to urinary tract injury and scarring, ultimately leading to end stage UTI. In a subtropical country like ours, there is a temporal relationship in the antibiotic sensitivity pattern of UTI. Hence, frequent large-scale studies are required from time to time to note the change in the sensitivity and resistance. Complicated UTI and subsequent renal failure continues to be one of the major causes of mortality in children. From this study, it can be concluded that *E.coli* still remains the commonest isolate in UTI. Nitrofurantoin, Amikacin, Norfloxacin, Gentamicin can be considered effective drugs. An emerging resistance has been noticed with Ceftazidime, Ofloxacin and Amikacin. Vancomycin may be reserve drug of choice in failed or multidrug resistance cases for gram-positive bacteria. Large-scale multicenter studies are required to generalize the data for the whole country.

I. Introduction

Urinary tract infection (UTI) is common in pediatric practice and an important cause of morbidity and mortality in children. However, UTI is a common problem throughout the world, the microbial isolates and their sensitivity pattern need to be analyzed at regular interval to monitor the changing pattern of microbial flora and the development of resistance to drugs, which may help the physician to treat UTI in better way and to prevent further complications.

II. Materials and Methods

We conducted the prospective analysis ofthe cases attending pediatric OPD and those admitted in the ward of Kanti Children's Hospital, Kathmandu, Nepal. Study period was six months from August 2012 to November 2012. Children of both sexes up to the age of 14 years were included in the study. Their clinical presentation with associated condition and risk factors were noted. Approximately 1890 urine samples were screened and 300 urine samples showed positive culture result. Parents were explained about the study and professional care was taken to collect the urine sample for routine culture and sensitivity by sterile technique. Urine was sampled for culture by aseptic supra pubic bladder aspiration in infants. Sterile plastic receptacles were used for collection of urine in younger patients to avoid contamination with stool. Clean catch mid-stream urine was sampled in older children and

Author α σ: St. Xavier's College, Maitighar, Kathmandu.
e-mail: biduraryalmb7@gmail.com
Author ρ: Kanti Children's Hospital, Maharajgung, Kathmandu.

adolescents after proper cleansing of urethra and under supervision. The samples were than processed for routine microscopy. Only samples with more than 5 WBC per high power field (hpf) were subjected for culture and antimicrobial susceptibility testing in the bacteriology laboratory of Kanti Children Hospital. Receptacle sample and mid-stream urine sample with culture with $>10^5$ colony forming units of bacteria/ml of urine in young infants and adolescents. Any colony count with supra pubic count in infants.

III. Observation and Result

Among the 1890 urine samples included in the study, 300(15.88%) showed positive culture result (fig. 1). Since, the study includes newly born babies up to 14 years of age. The high frequency of UTI was found in 0-2 years of age followed by 8-10 years of age. Among the 300 culture positive cases, 114(38%) were males and 186(62%) were females (Table 1).

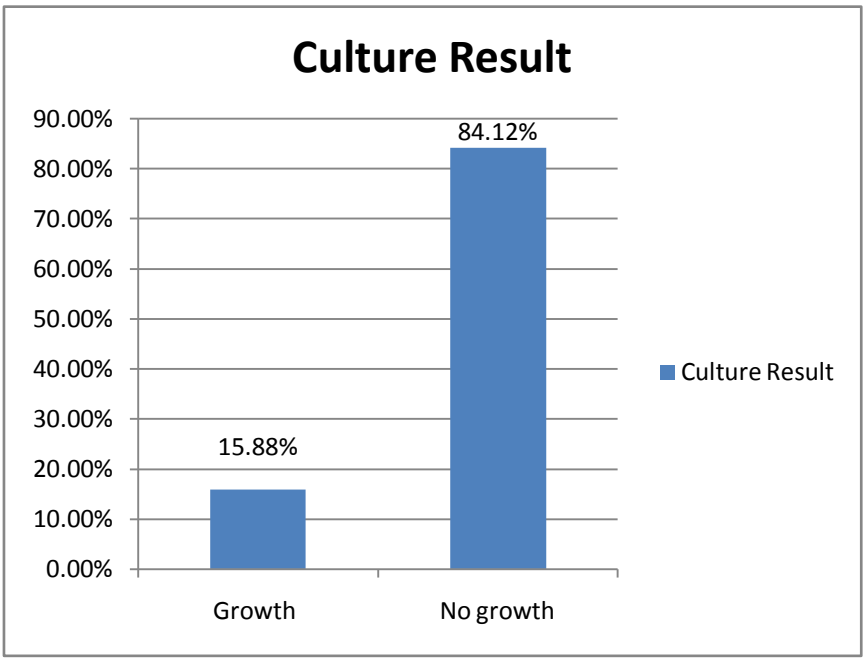

Figure 1 : Urine Culture Report

Table 1 : Age and sex distribution at Presentation (N=300)

Sex	Age(years)						
	0-2	2-4	4-6	6-8	8-10	10-12	12-14
Male	62	17	10	7	8	6	4
Female	90	31	25	13	10	10	7
Total	152	48	35	20	18	16	11

(14%) followed by Streptococcus fecalis (0.66%) of cases. (fig 2)

Among the gram negative isolates, the most common organisms isolated was *E. coli*(52.33%)followed by *Klebsiella pneumonia* (16%), *Pseudomonas aeruginosa* (4.33%), *Proteus mirabilis* (2.66%), *Citrobacter freundii* (2%), *Acinetobacter spp.*(1.66%), *Klebsiella oxytoca*, *Enterobacter spp.*, *Hafniaalvei* (1.33%), *Proteus vulgaris*, *Citrobacter koseri* (1%) and *Salmonella paratyphi* B (0.33%). *Staphylococcus aureus* (14%) followed by *Streptococcus fecalis* (0.66%) of cases. (fig 2).

Among the antibiotics used, Nitrofurantoin was found to have the highest sensitivity (71.67%) amongst most bacteria. Amikacin, Norfloxacin and Gentamicin had sensitivity of 69%, 61.71% and 61.67% respectively. *Pseudomonas aeruginosa* was 100% sensitive to Tobramycin, Piperacillin and Imipenem. Though sensitivity to Vancomycin was tested to 44 cases and it showed no resistance and it was 100% sensitive to *Staphylococcus aureus* and *Streptococcus fecalis*. Highest degree of resistance was noted with Ceftazidime(64%), Ofloxacin(61.33%), Ampicillin(60%) , ciprofloxacin (55.67%), Cotrimoxazole (52%), Gentamicin (38.33%), Amikacin (28%) and Nitrofurantoin (23.67%). The sensitivity pattern of various organisms was also studied. *E.coli* responded better with Nitrofurantoin, Aminoglycosides and Fluroquinolones but displayed a highresistance with most of thebeta lactams. Resitance was also noted with Ofloxacin, Nalidixic acid and Ciprofloxacin.

The sensitivity pattern of *Klebsiella* and *Proteus* was similar with few minor differences like *Klebsiella*showing high sensitivity with Amikacin (77.09%), Nitrofurantoin (70.83%), Norfloxacin (68.75%), Ceftazidime (64.58%) as compared to *Proteusspp* showing sensitivity to Amikacin, Norfloxacin, Ciprofloxacin (62.5%), Ofloxacin, Ceftazidime (37.5%). Amongst the gram negative bacteria *P. aeruginosa*was 100% sensitive to Piperacillin, Imipenem and Tobramycin. Amikacin was 100% sensitive to *Citrobacterkoseri*, *Enterobacterspp.*, *Salmonella paratyphi* B.*Citrobacterfreundii*, *Acinetobacter spp.*, was sensitive to Amikacin. *Hafniaalvei* was 100% sensitive to Nalidixic acid.

*Klebsiella spp.*and *Proteus spp.* showed high degree of resistance with beta lactams, Fluoroquinolones, Sulfonamides and Nitrofurantoin. *Salmonella paratyphi* B was 100% resistant to Fluroqouinolone, Aminoglygosides and Nitrofurantoin. *Staphylococcus aureus*and *Streptococcus fecalis*s-howed high sensitivity to Sulphonamides, beta-lactams and Nitrofurantoin whereas similar type of resistance with beta-lactams and Fluroquinolone.

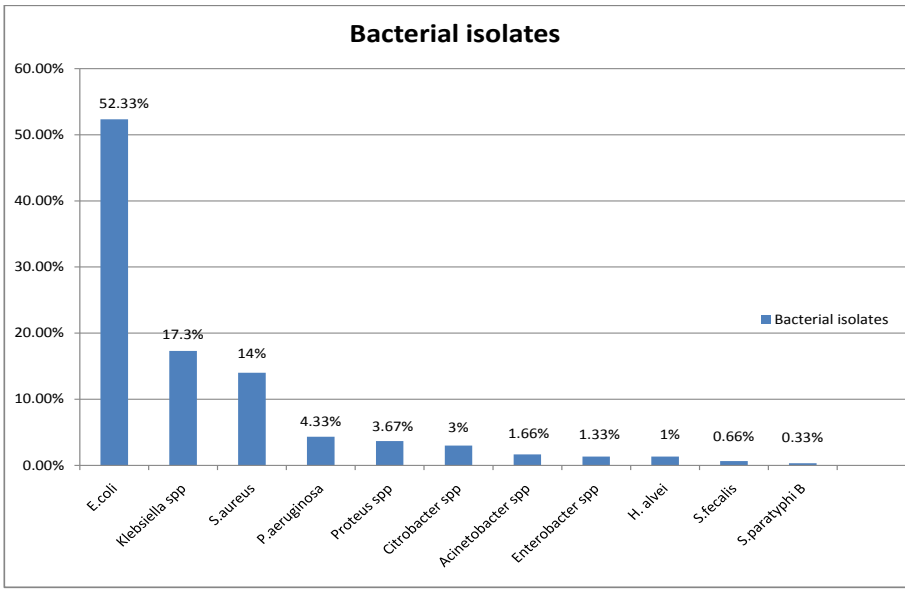

Figure 2 : Bacterial isolates of culture positive cases

Table 2 : Resistant Pattern of Bacterial Isolates Against Commonly used Antibiotics and Observation of MDR Bacterial Isolates

Organisms	Total no. of isolate	Resistant to				
		0 Drug (%)	1 Drug (%)	2 Drug (%)	MDR strains	
					>2drug	%
E. *coli*	157	9(5.73)	14(8.91)	29(18.47)	105	66.87
K. *pneumonia*	48	5(10.4)	3(6.25)	13(27.08)	27	56.25
K. *oxytoca*	4	0(0)	1(25)	1(25)	2	50
P. *mirabilis*	8	0(0)	2(25)	1(12.5)	5	62.5
P. *vulgaris*	3	0(0)	0(0)	1(33.34)	2	75
P. *aeruginosa*	13	0(0)	0(0)	0(0)	13	100
C. *koseri*	3	0(0)	0(0)	1(33.34)	2	75
C.*fruendii*	6	0(0)	1(16.67)	2(33.34)	3	50
Acinetobacterspp	5	1(20)	0(0)	0(0)	4	80
Enterobacterspp	4	0(0)	0(0)	1(25)	3	75
S. *aureus*	42	0(0)	2(4.76)	5(11.90)	35	83.33
S. *fecalis*	2	0(0)	0(0)	0(0)	2	100
H. *alvei*	4	0(0)	1(25)	0(0)	3	75
S. *paratyphi B*	1	0(0)	0(0)	0(0)	1	100
Total	**300**	**15(5)**	**24(8)**	**54(18)**	**207**	**69**

IV. Discussion

Urinary tract infection (UTI) is common cause of febrile illness in young children. In the first year of life. Urinary tract infection(UTI) is one of the most important causes of morbidity in the general population and the second most common cause of hospital visits(Das et al., 2006). Urinary tract infection(UTI) is not uncommon cause of bacterial illness in children, 4-8% of children have had an UTI from a population-based study(Suresh kumaret al., 2009). The prevalence and incidence of is higher in female than in male children, which are likely the result of several clinical factors including anatomical differences, hormonal effects and behavior pattern(Griebling, 2009). The prevalence of UTIs is quite different between two gender and age with high incidence in girls (1% in male and 3% in female), except the male infants with an incidence of 0.7% compared to the 0.1-0.4% of female infants (Foxman, 2002), which is due to bacteria harboring in prepuce of young infants.

Among the growth positive samples, 144(48%) were male patients and 156(52%) were female patients. Among 1890 urine samples, 1094(57.88%) were symptomatic, in which 166(15.18%) was culture positive. Urinary symptoms like dysuria, burning urine, increased frequency, haematuria, oliguria, bed wetting, chills and rigors, abdominal pain, vomiting, loose stool, etc. The first and the most critical step in establishing the diagnosis of UTI in infants and young children is the method by which the urine is collected. In the young infants care must be taken to prepare carefully the perineum and periurethral area for placement of sterile plastic receptacle for collection of urine. In the infants, the purest way to obtain urine for culture aseptically is by precutaneoussuprapubic aspiration. Older children and adolescents can be instructed to collect the midstream urine specimen after proper cleansing of the urethral area. These steps were strictly followed for collection of urine in our study. The presence of 105 organisms or more per ml of urine is diagnostic of UTI. If 103 to 105 colony forming units of a single genus and species per ml are recovered from two successive urine culture of a child, a diagnosis of UTI should be made6.

In our context, such cases were not included in our study as it was difficult to call the patient for repeated urine culture though they were empirically treated as suspected UTI. The suprapubic bladder aspiration or by catheterization contain fewer than 105 organisms because the organisms have not had sufficient time to multiply before the removal of urine from the bladder(Griebling TL.,2009).

In this study, 300(15.88%) resulted a positive culture in urine with significant colony count of ≥105rest 1590(84.12%) were culture negative or they had colony count <105 (fig.1). E.coli(52.33%) was found to be predominant organism in this study which resembles with the study done by Raiet al., (2008); Maliangoet al., (2012); Beyene and Tsegaye (2011);Daoud and Afif (2011); Elkehiliet al., (2010); Aypaket al., (2009); Hawn et al., (2009). Our distribution of pathogens wereE.coli(52.33%)was predominant organism isolated followed by Klebsiella pneumonia (16%), Staphylococcus aureus (14%), Pseudomonas aeruginosa (4.33%), Proteus mirabilis (2.66%), Citrobacterfruendi (2%), Acinetobacterspp (1.66%), Klebsiellaoxytoca (1.33%), Enterobacterspp (1.33%), Hafniaalvei (1.33%), Proteus vulgaris (1%), Citrobacterkoseri (1%), Streptococcus fecalis (0.66%) and Salmonella paratyphi B (0.33%).E.coli is by far the most common bacteria isolated from urine samples in both outpatients and inpatients of both sexes in children. This finding is also in agreement with findings ofTanejaet al., (2010); where E.coli (47.1%), Klebsiellaspp(15.6%), Enterococcus fecalis(8.7%), Proteus spp(5.9%), P. aeruginosa(5.9%) and others 17.1%.Yet in another study, the findings were consistent with ours where the pathogens were E.coli (47%), Klebsiellaspp (18%), S.aureus (13.4%), Proteus spp (9%), E.fecalis (5.3%),P.aeruginosa (5%), and others 2.3%.

In our study,Nitrofurantoin was found to have the highest sensitivity (71.67%) amongst most bacteriawhereas Proteus, Salmonella paratyphi B and P. aeruginosa was resistant to the same.Amikacin, Norfloxacin and Gentamicin had sensitivity of 69%, 6

1.71% and 61.67% respectively. Pseudomonas aeruginosa was 100% sensitive to Tobramycin, Piperacillin and Imipenem. Though sensitivity to Vancomycin was tested to 44 cases it was 100% sensitive to Staphylococcus aureus and Streptococcus fecalis. Highest degree of resistance was noted with Ceftazidime (64%), Ofloxacin (61.33%), Ampicillin (60%), Ciprofloxacin (55.67%), Cotrimoxazole (52%), Gentamicin (38.33%), Amikacin(28%) and Nitrofurantoin (23.67%).

In this study, 69% (207/300) were found to be Multidrug resistant (MDR) i.e. they were resistant to more than two drugs which is similar to the result of Pokhrelet al., 2006 in which 60.40% were MDR. The MDR in E.coli was found to be 66.87% (105/300). Although multidrug resistance was shown 100% by P. aeruginosa,Enterococcus fecalis and S. parathypi B, these were low in number and considered insignificant. In a study done by Tuladharet al., 2003 at TUTH, MDR bacterial strains were detected in 35.2% cases in which the most predominant was E.coli(22.2%) followed by Klebsiellaspp (6.1%) and Staphylococcus aureus (2.2%).

V. Conclusion

As UTI is the significant problem in the children and still continues to be a major threat for morbidity and mortality in subtropical parts of the world, larger scale studies must be carried out at a regular intervals in order

to identify the changing trend in the pathogenic organisms and update on its changing antibiotic susceptibility. Based on the sensitivity patterns we recommend empirical use of Nitrofurantoin, Amikacin, Norfloxacin and Gentamicin for patients with UTI.Vancomycin showed 100% sensitivity to gram-positive bacteria. Gram-negative bacteria like Proteus spp, P. aeruginosa and S.paratyphi B was 100% resistant to Nitrofurantoin. P.aeruginosa was 100% resistant to Imipenem, Piperacilin and Tobramycin. So, Vancomycin should be kept as reserve drug for gram positive organisms and Tobramycin, Imipenem and Piperacilin for P. aeruginosa.

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