



Isolation and Identification of Pathogenic Bacteria from Urinary Tract Infections in Infants from Different Hospitals of District Peshawar

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Abstract

In the present study isolation and identification of pathogenic bacteria, causing urinary tract infection in infants was performed. The research work was carried out at the Microbiology Laboratory, Abasyn University, Peshawar. Samples were collected from different hospitals of District Peshawar. The collected samples were inoculated on different media for identification and isolation such as Nutrient agar, MacConkey agar, Mannitol salt agar and CLED media. For further identification Gram staining and Biochemical tests were performed such as Catalase, Oxidase, Coagulase, Indole and TSI. In total of 90 samples, *E. coli* was most prominent, which was found to be 52 (57.77%) followed by *S. saprophyticus* 22 (24.44%), *K. pneumonia* 11 (12.22%). While least prominent was *S. aureus* 5 (5.55%). Among the total isolates the percentage of Gram-negative bacteria in UTI patients was higher than Gram-positive bacteria. Whereas the percentage of *E. coli* was (53%) and *K. pneumonia* was (14%) while the percentage of *S. saprophyticus* was (28%) and *S. aureus* (5%). Among the total 90 isolates, the highest prevalence was observed in female infants about (58.88%) while in male infants, the percentage was (41.11%).

Key words: Urinary Tract Infection, Infants, Pathogenic Bacteria.

Article Info:

Received:
April 26, 2018
Received Revised:
May 22, 2018
Accepted:
May 23, 2018
Available online:
May 25, 2018

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How to cite:

Faridullah, Muhammad D, Ishaq MS, Zoreen A, Farooq S, Siddiqi UA. Isolation and identification of pathogenic bacteria from urinary tract infections in infants from different hospitals of district Peshawar. *Abasyn Journal of Life Sciences* 2018; 1(1): 19-25.

1. INTRODUCTION

The urinary system consists of two ureters, two kidneys, bladder, and urethra. Bacteria are not mostly found in these areas. An infection can develop when bacteria enter the kidneys or bladder. These infections are basically called urinary tract infections ¹.

A urinary tract infection (UTI) is a serious bacterial infection causing illness in children and infants. UTI is the combination of clinical features and bacterial presence in urine. UTI can also be defined as the presence of more than 100,000 CFU/mL of bacteria after culturing urine (despite of symptoms). Clinical symptoms of UTI usually include frequency, fever, abdominal pain, urgency, dysuria and back pain. Both urinalysis and urine culture are done for the diagnosis of Urinary Tract Infection. Mostly, UTI is caused gram-negative bacteria such as *Klebsiella spp* and *E. coli*, but in some cases gram-positive bacteria are also responsible for UTI ².

UTIs are categorized into either upper tract infection, located in the ureters, collecting system, and parenchyma and lower tract infection, located in the bladder or urethra (cystitis and urethritis). For accurate diagnosis, it is important to understand the difference between the two types of UTIs. An inflammatory condition of the urinary bladder is called cystitis, whereas a diffuse pyogenic infection of the pelvis and parenchyma of the kidney is called pyelonephritis. Signs and symptoms of cystitis include dysuria, haematuria, frequency, malodorous urine, urgency, suprapubic pain and enuresis. While the signs and symptoms of Pyelonephritis include; positive urine culture, fever more than 38.5 °C, chills with flank pain and tenderness with pyuria. Pyelonephritis shows the most severe type of UTI in children with irreversible damage and a great morbidity rate. UTI symptoms include back pain, abdominal pain, dysuria, frequency, new-onset incontinence, but not a single symptom alone is sufficient to diagnose UTI in verbal children³.

UTI is one of the most common childhood infections, affecting round 8.4% of girls and 1.7% of boys by the age of 1-24 months. A third to one half of affected children will suffer from at least one recurrence. Gram negative bacteria such as *Proteus*, *Escherichia coli*, *Pseudomonas*, *Klebsiella*, *Enterobacter* and *Serratia* species are mostly involved in causing UTI. In 15%-65% children affected with UTI, permanent renal cortical scarring may occur specially in recurrent UTI and its long-term complications may include hypertension and chronic renal failure which results in end stage renal diseases. To prevent long-term damage with recurrent UTI several interventions have been tried which aim to reduce one or more of the factors that facilitate the development of UTI^{4,5}.

Urinary tract infections (UTI), if not treated in the beginning, cause high morbidity and long term complications in children. The incidence varies according to races, age and sex of children. Urinary Tract Infection (25.6%) is more common in male children under the age of one year. UTI occurs in 3-5% of girls and 1% of boys. It is noticed that 2% of boys and 7% of girls will remain asymptomatic, culture confirmed UTI by 6 years of age. In infants, symptoms of UTI can be non-specific. The prevalence of UTI in febrile infants is greater with younger age, with a range of nearly 7% among the febrile new-born^{2,6}.

Pediatric UTI's mostly account for 0.7% of physician office visits and 5–14% of emergency department visits by children annually. Diagnosis of UTI accurately is an important clinical implications; mostly febrile infants with UTI show indication of pyelonephritis. Nevertheless, the symptoms of UTI in childhood are nonspecific especially in infants, definitive testing for UTI involves bladder catheterization. Clinicians mostly are stucked with the decision of whether to obtain a urine sample for culture and urinalysis or not⁷.

UTI is among one of the most common infections, therefore the present study was conducted to isolate and identify the pathogenic bacteria causing urinary tract infections in infants.

2. MATERIALS AND METHODS

2.1 Collection of Sample

Ninety urine samples were collected from the patients from LRH, KTH, HMC, Al Khidmat Hospital and private clinical laboratories using sterile urine bags and bottles. Samples were transported to the Microbiology Laboratory at Abasyn University, Peshawar for culturing and identification of the microorganisms.

2.2 Culture Media Used

Different media were used for the culturing of samples i.e. Nutrient Agar, MacConkey Agar Media, Blood Agar, Mannitol Salt Agar and CLED Agar. CLED agar is a differential non-selective media used for culturing of bacteria present in urinary tract. CLED is used for differentiation and identification of all urinary pathogens, and many contaminants such as lactobacilli, diphtheroids and micrococci⁸.

2.3 Identification of bacteria

Gram staining was performed for the identification of bacteria i-e to differentiate between Gram positive and Gram negative bacteria.

2.4 Confirmation of bacteria

Different bio-chemical tests were applied for the confirmation of bacterial isolates like Catalase, Oxidase, Coagulase, TSI (triple sugar iron), Urease, Indole and Citrate.

3. RESULTS AND DISCUSSIONS

In the present study, 90 urine samples were screened for the presence of various bacterial pathogens. Out of the tested samples, different pathogens were identified, isolated and characterized. Fig. 1 shows that *E. coli* was the most pre-dominant isolated specie i-e 52 (57.77%) while 22 (24.44%) samples were positive for *S. saprophyticus*, 11 (12.22%) for *K. pneumoniae*. The least prevalent bacterial specie detected was *S. aureus* i-e 5 (5.55%).

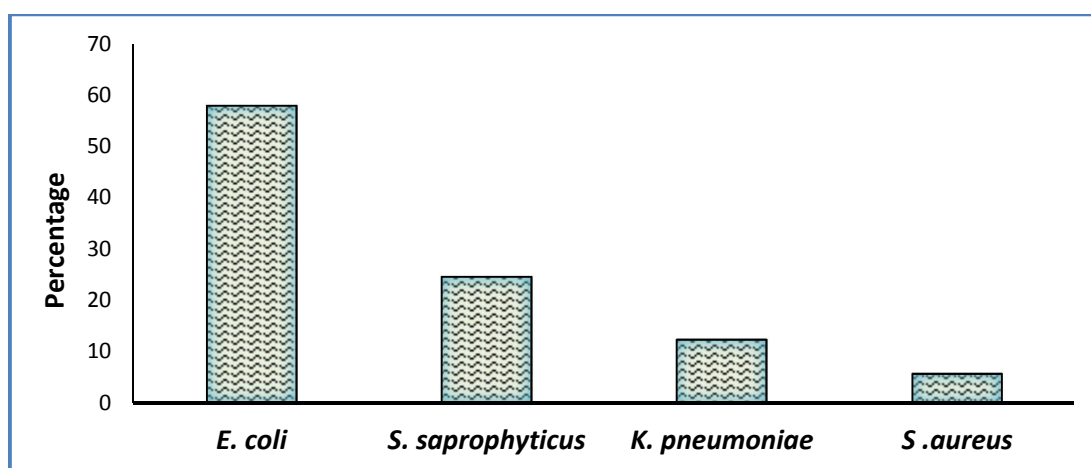


Fig. 1. Percentage wise distribution of various pathogenic bacteria isolated in urine specimens.

The data in Table 1 indicated biochemical tests results of different isolated bacterial samples. According to the results *S. aureus* demonstrated positive reactions towards Gram staining, Urease, Coagulase and Citrate and negative reactions towards Oxidase, Indole and Motility. While *S. saprophyticus* demonstrated positive reaction towards Gram staining, Urease, Catalase, Indole and negative reaction towards Motility, Citrate, Coagulase and Oxidase. *E. coli* showed positive reactions towards Catalase and Indole while it was negative for all the rest of biochemical tests carried out during the research work. While *K. pneumoniae* showed positive reactions towards Catalase, Urease, and Citrate and negative reactions towards Urease, Oxidase, Motility, Coagulase and Indole tests.

Table 1. Biochemical Tests carried out for identification of the Bacterial species Isolated, (+) represents presence and (-) represents absence.

Name of Bacteria	G. S	Cat	Oxi	Ure	Cit	Ind	Mot	Coag
<i>E. coli</i>	-	+	-	-	-	+	-	-
<i>S. saprophyticus</i>	+	+	-	+	-	+	-	-
<i>K. pneumoniae</i>	-	+	-	+	+	-	-	-
<i>S. aureus</i>	+	+	-	+	+	-	-	+

Key: G. S = Gram staining; Cat = Catalase; Oxi = Oxidase; Ure = Urease; Cit = Citrate; Ind = Indole; Mot = Motility; Coag = Coagulase, + = Positive; - = Negative

The data in Table 2 specified following biochemical results of TSI Agar for *S. aureus* displayed overall negative reactions towards Slant, Butt, CO₂ and H₂S while *S. saprophyticus* also shown overall negative reactions towards Slant, Butt, CO₂ and H₂S. *E. coli* presented positive reactions towards Slant, Butt, CO₂ and negative to H₂S while *K. pneumoniae* shown positive reactions towards Slant, Butt and as well CO₂ and H₂S negative.

Table 2. Response/assessment of isolated/test organisms towards TSI

Name of Bacteria	Triple Sugar Iron Reaction			
	Slant	Butt	CO ₂	H ₂ S
<i>S. aureus</i>	-	-	-	-
<i>S. saprophyticus</i>	-	-	-	-
<i>E. coli</i>	Acidic	Acidic	+	-
<i>K. pneumoniae</i>	Acidic	Acidic	+	-

Key: + = Positive; - = Negative

Among the total isolates the percentage of Gram negative bacteria in UTI patients was higher than Gram positive bacteria. The frequency of *E. coli* was 52 (57.77%) and *K. pneumoniae* was 11 (12.22%) while the percentage of *S. saprophyticus* was 22 (24.44%) and *S. aureus* was 5 (5.55%). The data regarding the prevalence of various bacterial species is shown in Fig. 2.

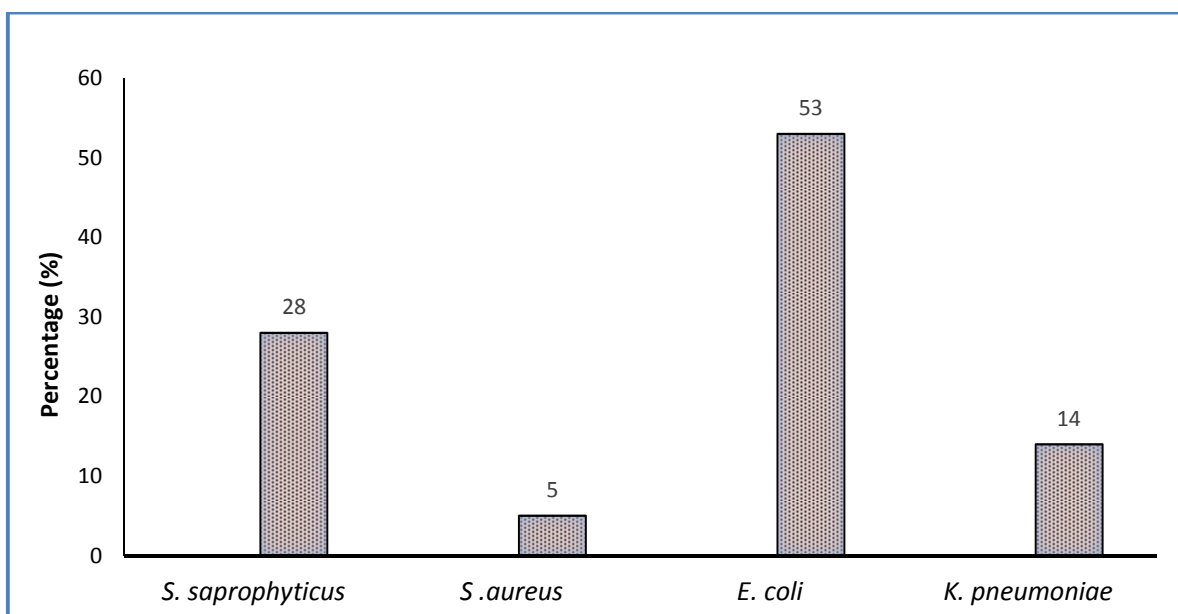


Fig. 2. Frequency distribution among uropathogens.

The prevalence of uropathogens among positive patients by sex is shown in Fig. 3. Among the total 90 isolates, the highest prevalence was observed in female infants about (58.88%) while in male infants the percentage was (41.11%).

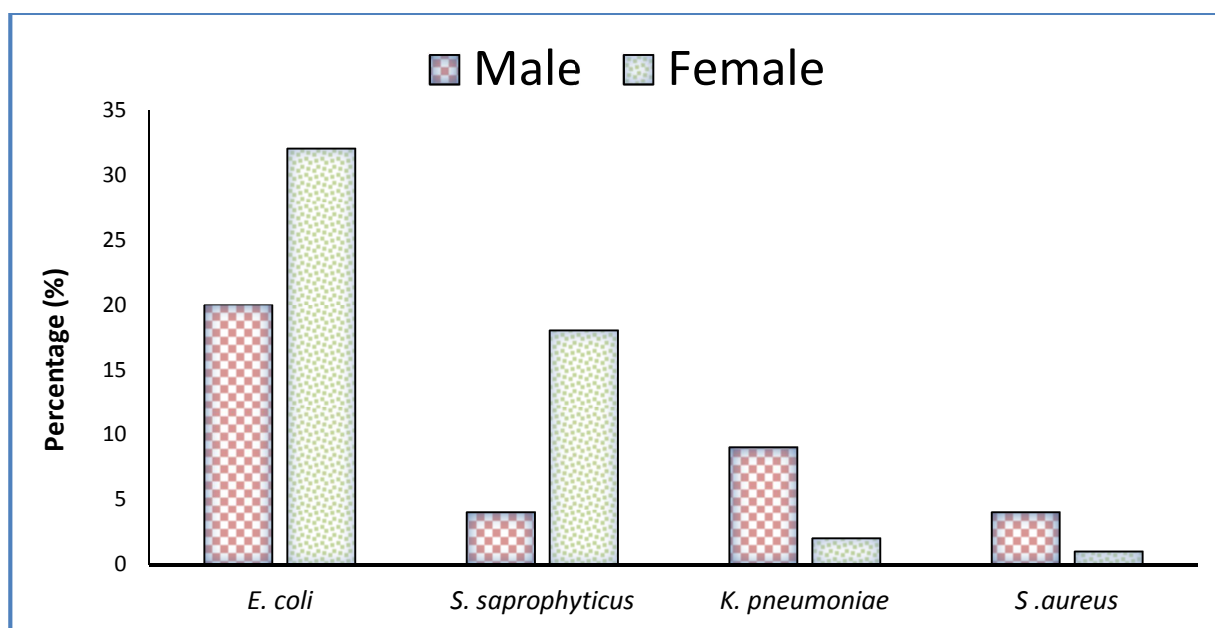


Fig. 3. Prevalence of uropathogens among positive patients by sex.

UTI is a major clinical problem in infants as (especially those who are febrile) the infection usually consists of acute pyelonephritis, which affects the renal function adversely later in life ⁸.

In a systematic review, almost 7% of Infants aged between 2 to 14 months having fever and without any major urinary symptoms were diagnosed with a Urinary Tract Infection. Depending on sex, age and race the occurrence rate varies. The rate in uncircumcised febrile boys <3 months of age was 20.7% compared with 2.4% in circumcised boys, declining to 7.3% and 0.3%, respectively, in boys six to 12 months of age. In febrile girls, approximately 7.5% < 3 months of age, 5.7% three to six months of age, 8.3% six to 12 months of age and 2.1% 12 to 24 months of age had a UTI as the cause of their fever. Contamination is a major issue in obtaining a urine sample from a male when the foreskin cannot be retracted and the rates in uncircumcised males are, undoubtedly, overestimates ⁹.

Among the total 90 isolates, the highest prevalence was observed in female infants about (58.88%) while in male infants the percentage was (41.11%) while Paintsil *et al.*,¹⁰ also shown the same prevalence in UTI, affecting round 1.7% of boys and 8.4% of girls. A third to one half of affected children will suffer from at least one recurrence, it is usually occurs as an ascending infection in older children, where the common organism involved include Gram negative bacteria such as *E. coli*, *Klebsiella*, *Proteus*, *Enterobacter*, *Pseudomonas* and *Serratia* species and Gram positive bacteria such as *S. saprophyticus* and *S. aureus*.

In the present study, among the isolates, 52 (57.77%) were detected *E. coli*, which was the most prevalent bacterial specie while 22 (24.44%) samples were positive for *S. saprophyticus*, 11 (12.22%) for *K. pneumoniae*. The least prevalent bacterial specie detected was *S. aureus* which was found 5 (5.55%) while Ogbukagu *et al.*¹¹ concluded that *E. coli* (24.2%), *Klebsiella* spp. (18.2%), *S. aureus* (18.2%), *Proteus mirabilis* (9.1%), *Pseudomonas aeruginosa* (9.1%), *Enterococcus faecalis* (9.1%), *Citrobacter intermedium* (6.1%) and *S. saprophyticus* (6.1%) which showed minute distinction in isolated pathogens due to species variations and geographical distribution.

Among the isolates, 52 (57.77%) were detected *E. coli*, which was the most prevalent bacterial specie while 22 (24.44%) samples were positive for *S. saprophyticus*, 11 (12.22%) for *K. pneumoniae*. The least prevalent bacterial specie detected was *S. aureus* which was found 5 (5.55%) while Kalantar *et al.*¹² demonstrated the isolates which were *E. coli*, *Klebsiella*, *Proteus*, *Enterobacter*, *Pseudomonas* and *Serratia* species which showed variation to our research due to geographical differences.

According to results the percentage of Gram negative bacteria in UTI patients was higher than Gram positive bacteria. The frequency of *E. coli* was 52 (57.77%) and *K. pneumonia* was 11 (12.22%) while the percentage of *S. saprophyticus* was 22 (24.44%) and *S. aureus* was 5 (5.55%) while Sawalha et al. ² also verified that Gram negative bacteria were responsible for 59.25% of UTIs in comparison to gram positive bacteria which were 40.7%. *E. coli* was the most predominant uropathogens with (51.8%) followed by *S. aureus* (29.6%) which showed conformity to our analysis.

4. CONCLUSIONS

The study concluded that the prevalence of *E. coli* was higher in all urine samples, which was found to be 52 (57.77%) followed by *S. saprophyticus* 22 (24.44%), *K. pneumonia* 11 (12.22%) and *S. aureus* 5 (5.55%). It was also concluded that the prevalence of Gram negative bacteria in UTI patients was higher than Gram positive bacteria, in which the percentage of *E. coli* was 53% and *K. pneumonia* was 14% while the percentage of *S. saprophyticus* was 28% and *S. aureus* was 5%. Among the total 90 isolates, the highest prevalence was observed in female infants (58.88%) while in male infants the percentage was (41.11%).

ACKNOWLEDGEMENTS

The authors thank the management staffs of LRH, KTH, HMC, Al Khidmat Hospital and other private clinical laboratories for their helps in sample collection.

CONFLICT OF INTEREST

All authors declare no conflict of interest regarding this article.

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