

Rising Incidence of Streptococcal Urinary Tract Infections: A Retrospective Clinical and Microbiological Assessment

Farhan Rasheed, Ahmad Yar*, Raja Kamran Afzal**, Muhammad Bilal***, Iqra Jamill****, Asma Ejaz

Department of Pathology, Allama Iqbal Medical College, Lahore, Pakistan, *Department of Pathology, Combined Military Hospital Sialkot/National University of Medical Sciences (NUMS) Pakistan, **Department of Pathology, Armed Forces Institute of Cardiology/National Institute of Heart Diseases, Rawalpindi/National University of Medical Sciences (NUMS) Pakistan, ***Department of Pathology, Nawaz Sharif Medical College, University of Gujrat, Gujrat Pakistan, ****Department of Microbiology, University of Central Punjab, Lahore Pakistan

ABSTRACT

Objective: To determine distinct species of Streptococci along with their antimicrobial susceptibility pattern in urine samples.

Study Design: Retrospective Analytical Study.

Place and Duration of Study: Department of Pathology, Allama Iqbal Medical College, Lahore, Pakistan, from Jan 21 to Mar 2023.

Methodology: Urine samples of included subjects were processed according to standard microbiological procedures. Microscopy and culturing of urine were done. Organisms' identification was done by different biochemical tests. AST was performed by the disc diffusion method according to CLSI guidelines.

Results: Out of 7346 urine samples, 2377(32.4%) yielded growth. E. coli was most prevalent and yielded 957 (63.5%) positive results. Among 829 Streptococci spp., Enterococcus faecalis 492(59.3%) yields the highest number. The most effective antibiotics were found to be Vancomycin, Teicoplanin, and Nitrofurantoin.

Conclusion: Unusual urinary isolates, including distinct streptococcal species, should be carefully monitored. Nitrofurantoin and Ceftriaxone showed high susceptibility rates and may be suitable for empirical UTI treatment.

Keywords: Antimicrobial susceptibility testing, Clinical Laboratory and Standards Institute, Urinary tract infections.

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INTRODUCTION

Urinary tract infections (UTIs) are considered the most prevalent bacterial infections, and about 150 million to 250 million cases are reported annually across the world. By 2011, the cost of UTI-associated hospitalization per year in the USA had reached US \$9.7 billion.¹ Clinically, UTI presentations may be variable and can range from simple cystitis to complicated urosepsis. The pattern of microorganisms causing UTIs may vary from region to region.² Proper antibiotics with consistent effectiveness are vital for the treatment.³ Nevertheless, some bacteria have developed the ability to survive in an antibacterial drug environment, which eventually results in "antibiotic resistance. Frequent urination (or an urge to urinate) and burning sensation during urination without vaginal discharge and substantial pain are considered common symptoms of UTIs. Urine cultures are necessary for the diagnosis of UTIs. After isolation of microorganism, sensitivity testing is usually done by dilution testing. The most common urinary

pathogens are Gram-negative rods, including the Enterobacteriaceae family and Pseudomonas Aeruginosa, and Gram-positive cocci, including enterococci, along with Candida spp. accounting for 80% of the community-associated infections and 40% of the nosocomial infections.⁴ Epidemiological data and the sensitivity profile of uropathies are crucial for empirical antibiotic treatment determination.⁵ In the interim, there is a tremendous increase in antibiotic resistance across the world. Reasons for this increasing resistance are overuse, inopportune prescribing, the availability of a few new antibiotics, overuse and misuse of antibiotics, and regulatory barriers.⁶ Hence, it is a matter of hours to execute antibiotic-supervised programs effectively to minimize the likelihood of bacterial resistance.⁷ Antibiotic resistance is more common in nosocomial settings. But it is also spreading in community-acquired UTIs, and gram-positive cocci are becoming more resistant to commonly used antibiotics.⁸

Therefore, the present study was designed to determine the frequency of different Streptococci species isolated from urine samples and assess their antimicrobial susceptibility patterns in UTIs.

Correspondence: Dr Farhan Rasheed, Department of Pathology, Allama Iqbal Medical College, Lahore, Pakistan

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Identifying these organisms and local susceptibility trends may guide empirical therapy, reduce reliance on antibiotics, and reduce emerging resistance.

METHODOLOGY

The retrospective analytical study was done at the Pathology Department of Allama Iqbal Medical College, Lahore, from Jan 2021 to Mar 23. Ethical approval was obtained from the Ethical Review Board (ERB) of Allama Iqbal Medical College/Jinnah Hospital, Lahore (Ref No: ERB142/9/18-04-2023/S1 ERB, Dated: 18/04/2023). Sample size was calculated using the EPI software version 4.50 calculator, with a 95% confidence level, a 5% margin of error, and a reported prevalence of Streptococcal species in urine samples of 14.3%.⁹ The estimated sample size came out to be 114 urine samples. A non-probability purposive sampling technique was used.

Inclusion criteria: Culture-positive urine samples for Streptococcus species were included in patients of either gender and aged between 18 and 60 years with complaints of UTI.

Exclusion criteria: All other clinical samples other than urine samples were excluded. Samples from the same patient during same episode of illness and isolation of any other microorganism except Streptococcus species were not included in the study.

Mid-stream urine samples will be collected using sterile, wide-mouthed containers with screw cap tops and sent to Microbiology Lab for microscopy and culture. Microscopy was done on deposits of urine samples after centrifugation. After microscopy, urine samples were inoculated on Blood and CLED agar. After overnight incubation at 37 °C, identification of the microorganisms was done by Gram staining, biochemical tests, and serology. Confirmation of Streptococcus species was done by Lancefield grouping, beta hemolytic properties, Bacitracin disc, Bile esculin hydrolysis, 6.5% NaCl growth, and Vitek 2. 105cfu/ml was taken as a significant bacterial count. Antibacterial susceptibility of the isolates was determined by Kirby-Bauer disk diffusion method, and zones of inhibition were measured according to CLSI 2021 protocols.

Data was analyzed for Gender, Streptococci species, and their resistance pattern against different drugs were taken as study variables. Frequencies and percentages were calculated to estimate the antibiotic sensitivity pattern of distinct species against various drugs.

RESULTS

Out of 7346 urine samples, 2377(32.4%) yielded growth. Gender distribution showed that females were more commonly affected than males. Among 2377 total positive cases, 1687(71.0%) were females, while 690 (29.0%) were males.

Out of 2377, 1501(63.1%) were Gram-negative, while 876(36.9%) were Gram-positive organisms. Among gram-negative organisms, *E. coli* was most prevalent, 953(63.5%), followed by *K. pneumoniae*, 312(20.7%), and *P. aeruginosa*, 88(5.7%). Among Gram-positive organisms, *E. faecalis* was found to be most prevalent, 490(55.9%), followed by non-typeable streptococci species, 260(29.9%), and *S. agalactiae*, 41(4.9%). Distribution of distinct species of Streptococci is shown in Table-I.

Table-I: Distribution of Different Streptococci Species in Urine Samples (n=2377)

Organisms	Frequency (%)
Enterococcus Faecalis	492(59.3%)
Non-Typeable Streptococci Spp	263(31.8%)
Streptococcus Agalactiae	45(5.5%)
Streptococcus Mitis	9(1.1%)
Streptococcus Pyogenes	7(0.8%)
Enterococcus Faecium	6(0.7%)
Streptococcus Pneumoniae	4(0.5%)
Group F Streptococci	2(0.2%)
Group C Streptococci	1(0.1%)

Antibiotic susceptibility pattern of different Streptococci species against multiple classes of antibiotics is shown in Table-II.

DISCUSSION

In the present study, the prevalence of urinary tract infections was found to be 32.4%. Comparable results were reported in 2022, when a 32.2% positivity ratio of urinary tract infections was observed.¹⁰ UTIs are considered one of the most common bacterial infections in humans. Treatment of urinary infections is usually based on experience and selection of the most appropriate antibiotic for the disease, along with knowledge of trends in antibiotic resistance in that specific region. Another study reported a 31.8% prevalence of urinary tract infections. These results were in accordance with our results. A high ratio of negative cultures may be because of one or more reasons, including diagnostic inefficiency or improper sample collection.¹¹ Intermittent observations of the uropathogens along with their susceptibility resistance profiles in each region are necessary.

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Table-II: Resistance Pattern of Streptococcus Species Against Different Antibiotics

Antibiotics		E. faecalis n=492	Non-typeable streptococci spp n=263	S. agalactiae n=45	S. mitis n=9	S. pyogenes n=7	S. pneumoniae n=4
Aminopenicillin	Ampicillin	103(21.0%)	15(5.8%)	2(4.9%)	1(12.5%)	1(16.7%)	1(25.0%)
	Penicillin	108(21.9%)	13(5.0%)	2(4.9%)	1(12.5%)	1(16.7%)	1(25.0%)
Cephalosporins	Cefixime	-	92(35.0%)	11(24.4%)	7(75.0%)	2(33.3%)	3(75.0%)
	Ceftriaxone	-	12(4.7%)	1(2.4%)	0(0.0%)	1(16.7%)	1(25.0%)
Fluoroquinolones	Levofloxacin	396(80.6%)	161(61.2%)	17(39.0%)	7(75.0%)	4(66.7%)	2(50.0%)
Glycopeptides	Teicoplanin	2(0.3%)	3(1.1%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)
	Vancomycin	0(0%)	3(1.1%)	0(0.0%)	0(0.0%)	0(0.0%)	0(0.0%)
Misc Agents	Fosfomycin	77(15.8%)	52(19.7%)	2(4.9%)	2(25.0%)	1(16.7%)	2(50.0%)
	Linezolid	2(0.4%)	4(1.5%)	0(0.0%)	0(0.0%)	0(0.0%)	1(25.0%)
	Nitrofurantoin	75(15.2%)	18(7.0%)	2(4.9%)	1(12.5%)	2(33.3%)	1(25.0%)
Tetracyclines	Doxycycline	254(51.7%)	42(16.0%)	19(41.5%)	3(37.5%)	1(16.7%)	2(50.0%)

In our study, females were found to be more affected by UTI than males, with a percentage of 71% and 29%, respectively. Our results were in accordance with the findings of Muhammad *et al.*, which reported a 77.8% positivity UTI ratio in females.¹² Gram-negative microorganisms are considered the most common causative agents of urinary tract infection, but in our study, many of the gram-positive organisms are also found in urinary tract infections, especially Staphylococcus species, Enterococcus species, and different Streptococci species. Among the Streptococcus species, the most common is Group-B Streptococcus.¹³ It is exceedingly rare to find or isolate Streptococcus pyogenes from urine. In case of its isolation, clinical importance should be evaluated carefully.

In our study, the most common organisms among streptococci species were found to be Enterococcus faecalis, which is compatible with the study of Orji *et al.*, which reported Enterococcus faecalis as the most common organism among gram-positive organisms causing UTI, and also reported 4.5% S. agalactiae among organisms causing UTI.¹⁴ In the current study, the frequency of S. pyogenes was found to be 0.8% among different Streptococci species. A study by Mahdi *et al.*, reported a 2.6% prevalence of S. pyogenes, which was like our results.¹⁵ Frequency of S. agalactiae was found to be 5.5% in our study. Regarding antibiotic therapy, Streptococcus sp. is susceptible to many commonly used antibiotics, with teicoplanin, vancomycin, and nitrofurantoin showing maximum sensitivity against distinct species of streptococci.⁴

Many prior studies also reported comparable results to our study, in which nitrofurantoin showed the best action against most of the isolates. Comparing the results with the findings of the study conducted by Tandogdu *et al.*, 20% resistance against Nitrofurantoin was reported.⁵ In our study, Enterococcus faecalis

showed maximum sensitivity against Ampicillin and Penicillin, which is in accordance with the study of Bakhtiari *et al.*, in which Enterococcus species were most susceptible to penicillin and ampicillin.¹⁶ In our study, fluoroquinolones were found to be the least susceptible antibiotics. Consistent with our results, of Nolina *et al.*, who reported the highest resistance of fluoroquinolones against UTI pathogens.¹⁷ Ghaima *et al.*, reported comparable results.¹⁸ A Point mutation in DNA gyrase subunits can lead to this pattern of resistance.¹⁹

Isolation of unusual organisms in Urinary tract infections leads to the manifestation of an underlying uropathological illness, and microbiologists should emphasize the clinical history of the patient for the purpose of determining the significance of the uropathological illness. Furthermore, routine cultural practices of urinary samples in laboratories can lead to increasing probability of missing fastidious organisms. For this reason, the actual burden of the disease cannot be estimated properly. Moreover, overuse and misuse of given antibiotics by patients should be avoided to combat antibiotic resistance.

LIMITATIONS OF STUDY

This current study has some limitations. The retrospective design, single-center study, which may have caused limited geographic sampling, were downsides. In addition, inadequate clinical information about the patients and the inability to compare with other hospitals were other limitations.

CONCLUSION

In urinary tract infections, gram-positive organisms can also play a significant role and cause serious infections, Streptococcus pyogenes being one of them. Thus, the prospect of infection with such unusual organisms should be carefully considered, especially in cases of complicated urinary tract infections. Moreover, Nitrofurantoin and ceftriaxone have high susceptibility rates and are applicable in the empirical treatment of UTI.

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Authors' Contribution

Following authors have made substantial contributions to the manuscript as under:

FR & AY: Data acquisition, data analysis, critical review, approval of the final version to be published.

RKA & MB: Study design, data interpretation, drafting the manuscript, critical review, approval of the final version to be published.

IJ & AE: Conception, data acquisition, drafting the manuscript, approval of the final version to be published.

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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