

# Original Article

DOI: 10.53389/RJAHS.2023020107

# Prevalence and Antimicrobial Sensitivity Pattern of Staphylococcus saprophyticus isolated from Urinary Tract Infection in Women

Samra Asghar\*, Sidra Batool, Munaza Sajjad, Mahnoor, Aroosha Imran, Rumaila Khizar

Institute of Microbiology, University of Agriculture, Faisalabad

#### Author's Contribution

SAconception and design, SB, MSData collection and analysis, M,SBStatistical analysis, SA Final submission and approved for publication

#### Article Info.

Received: May 19, 2023 Acceptance: June 2, 2023 Conflict of Interest: None Funding Sources: None

### Address of Correspondence

Samra Asghar Institute of Microbiology, University of Agriculture, Faisalabad samraasghar@gmail.com

#### ABSTRACT

Background: Staphylococci saprophyticus are Gram-positive, coagulase-negative, nonhemolytic cocci which cause urinary tract infections in young adult women.

Objective: The present study was aimed to investigate the prevalence and antimicrobial sensitivity pattern of S. saprophyticus isolated from young women with urinary tract infection.

Methodology: A total of 100 urine samples were collected from urinary tract infected women at Government General Hospital, Faisalabad. Samples were transported and proceeded in Microbiology laboratory of Riphah international university, Faisalabad. The isolated S. saprophyticus were identified by Gram staining, culture characteristics, biochemical tests and by antimicrobial (Novobiocin) sensitivity test by Modified Kirby Bauer disc diffusion method.

Results: Ninety-four (94) out of 100 urine samples revealed growth of gram positive bacteria, gram negative bacteria and fungi on different culture media. S. saprophyticus was recovered from 3 urine samples. All strains of S. saprophyticus (100%) were found sensitive to imipenem and amikacin. Out of 3, two isolate (66.6%) showed sensitivity against gentamicin and ciprofloxacin. However, all isolated S. saprophyticus were found resistant to pipemidic acid and nitrofurantoin. These findings contribute to the understanding of infections caused by S. saprophyticus and may aid in the selection of appropriate antibiotics for the treatment of urinary tract infections.

Keywords: Urinary tract infection, Staphylococcus saprophyticus, Prevalence, Antimicrobial sensitivity test.

## Introduction

Staphylococcus saprophyticus is Gram positive, non-hemolytic and coagulase negative cocci. In women of childbearing age, it is a leading cause of Urinary tract infection (UTI), which in turn can lead to acute pyelonephritis and urethritis. 1,2 Various virulence factors contribute to its pathogenesis. Enzymes in the cytoplasm of S. saprophyticus include Urease and D-serine, as well as proteins such as fibronectin binding autolysin, surface associated lipase, uro-adherence factor, and collagen binding serine-aspartate-repeat protein. Surface proteins bound to the cell wall of S. saprophyticus activate its hemagglutinin, which helps the bacteria connect to cells in the urinary tract. Because the urethras of women are shorter and closer to the body, they are more likely to get UTIs than men. 3, 4 S. saprophyticus has accumulated genetic determinants encoding strong resistance to heavy metals [5] and detoxification of uric acid and D-serine, S. saprophyticus is able to persist in hard and toxic

environments, which contributes to its effectiveness as a uropathogen.<sup>6</sup> Additionally, it has been reported that S. saprophyticus pathogenicity is linked to its ability to stick to uroepithelial cells, which is facilitated by adhesins, surface proteins, and biofilm development.7 When bacteria invade the bladder and its surrounding structures, it's called a urinary tract infection. It is common for bacteria to enter the urinary tract via the urethra. Urinary tract infections often present with dysuria, pain in the suprapubic region, and increased urination frequency. Kidney stones and the use of spermicide-containing diaphragms as a form of birth control can both significantly increase the likelihood of a urinary tract infection. Lower UTIs affect the lower urinary tract (cystitis) can be painful and uncomfortable. Symptoms of an upper UTI include fever, chills, back or side discomfort, nausea, vomiting, anxiety, disorientation, or agitation. If left untreated, upper UTIs pose a

serious risk of kidney damage or infection in the bloodstream.[8] Due to a lack of routine susceptibility testing, the state of surveillance for antibiotic resistance in S. saprophyticus is inadequately documented. The Clinical and Laboratory Standards Institute (CLSI) states that S. saprophyticus urine isolates should not be routinely tested for infections since infections are sensitive to antimicrobial agent concentrations in urine, which are often used to treat simple, acute UTIs.9 It has been shown that S. saprophyticus can improve its virulence and antibiotic tolerance by 100 to 1000 times when it forms biofilms. in contrast to non-biofilm-producing isolates.7 Several mechanisms, such as the encoding of antibiotic-resistant genes, the restriction of drugs, and the counteraction of host immunity, contribute to the biofilm's ability to transmit antibiotic resistance. 10 It is common practice to treat a UTI with a broad-

spectrum antibiotic without first conducting a culture and sensitivity test; this is done on an empirical basis. The rise of multi-resistance in bacteria is directly attributable to the global spread of antibiotic resistance, which in turn is caused by the careless and overuse of these drugs.<sup>11</sup> Nearly 25,000 Europeans die each year from complications of UTIs caused by multidrug-resistant (MDR) bacterial strains, according to a study by the European Survey of Antibiotic Consumption. 12

# Methodology

The study was conducted in compliance with ethical guidelines. The study protocol was approved by the institutional review board of Riphah International University, Faisalabad and Government General Hospital, Faisalabad.

This experimental types of study were done at the Department of Medical Laboratory Technology, Riphah International University, Faisalabad. The study was carried out from March, 2023 to April, 2023. Urine samples were collected from female patients either admitted in inpatient department or visited the outpatient department of Government General Hospital, Faisalabad. A total of 100 urine samples were collected from young females suffering from urinary tract infections. The inclusion criteria specify the target population, such as sexually active adult females (18 to 45 years old). Collection and transportation of the samples were done according to standard guidelines of national healthcare facilities. Midstream urine was collected. Once the urine samples were collected, it was ensured that the samples were properly labelled with the patient's name, date, age, and transported to Riphah Microbiology laboratory within 2 hours. The samples were cultured on Blood and MacConkey agar. On blood agar, S. saprophyticus colonies appear as bright yellow or white pigment. S. aureus displays a light to golden yellow pigment,

whereas S. epidermidis shows a white pigment. The typical appearance of Streptococcus colonies appears as domeshaped with a smooth or moist surface and clear margins.[13] The isolated bacteria were identified and confirmed by Gram staining, biochemical tests ncluding Catalase and Coagulase tests, and Novobiocin sensitivity testing by following standard protocols. 14

Antiboitic sensitivity test was performed by modified Kirby-Bauer disc diffusion method and the zone of inhibition was measured by Vernier calliper by following instructions of CLSI.15 The antibiotic discs used for S. saprophyticus were Tobramycin, Amikacin, Fosfomycin, Pipemidic acid, Imipenem, Ciprofloxacin, Gentamicin and Nitrofurantoin.

# Results

Out of 100 urine samples, 94 samples were culture positive. Out of these 94 samples, Gram positive bacteria were recovered from 12 samples. While growth of 78 samples were identified as Gram negative bacteria. Out of 12 Gram positive isolates, 7 were identified as Staphylococcus. Out of these 7 Staphylococci, 3 were identified as Coagulase positive (S. aureus) and 4 were identified as Coagulase Negative Staphylococcus species. Out of 4 Coagulase negative Staphylococcus isolates, 3 isolates showed resistance to novobiocin, which indicated S. saprophyticus. The percentage positivity of S. saprophyticus and other microbes isolated from urine samples is summarized in Table I.

Table I: Prevalence of different microbes isolated from urine sample. Number and percentages of Names of Inclotes

Names of Isolates	lsolates N(%)		
Gram negative rods	78(78)		
Staphylococcus saprophyticus	3(3)		
Staphylococcus aureus	3(3)		
Streptoccous	3(3)		
Bacillus species	2(2)		
Staphylococcus epidermitis	1(1)		
Candida	4 (4)		
Total	94(100)		

All strains of S. saprophyticus (100%) were found sensitive to imipenem and amikacin. Out of 3, two isolate (66.6%) showed sensitivity against gentamicin and ciprofloxacin. However, all isolated S. saprophyticus were found resistant to pipemidic acid and nitrofurantoin. It was observed that 2 out of 3 isolates showed resistance against more than three antibiotics. Antimicrobial sensitivity pattern of S. saprophyticus is given in Table II.

Table II: Antimicrobial sensitivity	pattern	of S.	saprophyticus
(n=3) isolated from urine sample.			

(11-5) isolated from drifte sample.			
Antibiotic	Sensitive N (%)	Resistant N(%)	
Imipenem	3(100.00)	0(00)	
Amikacin	3(100.00)	0(00)	
Gentamicin	2(66.66)	1(33.33)	
Tobramycin	2(66.66)	1(33.33)	
Ciprofloxacin	2(66.66)	1(33.33)	
Fosfomycin	1(33.33)	2(66.66)	
Nitrofurantoin	0(00)	3(100.00)	
Pipemidic acid	0(00)	3(100.00)	

# Discussion

The recent study was conducted at Microbiology laboratory of Riphah International University, Faisalabad, that provide valuable insights into the prevalence and antibiotic sensitivity of S. saprophyticus. S. saprophyticus was recovered from 3% of UTI infected women in accordance with prevalence rate (3%) of a previous study conducted in Saudi Arabia. 16 Similarly, a low prevalence rate (2%) of S. saprophticus from UTI infected women has been documented by a previous study conducted at Dhaka, Bangladesh. 16 A surveillance study conducted in Japan on uncomplicated urinary tract infections (UTIs) reported 5% prevalence of S. saprophyticus that is a little bit higher as compared to our study.17

In our study, total 94 urine samples were found positive for microbial growth. isolated. Out of 94, 78% were gram negatives rods and 3% were S. saprophyticus. In consistence with the present study, a survey of antimicrobial resistance in urinary tract pathogens, carried out in 252 community care centres in 17 countries reported that the most common uropathogens were E. coli (53.3%) followed by S. saprophyticus (2.5%) among 4,734 females included in the survey.<sup>18</sup> A low prevalence rate of 0.07% of S. saprophyticus among UTI infected females has been reported in an old study conducted at Canada.[19] In contrast, another studies conducted at Brazil and Bangladesh revealed high prevalence of S. saprophyticus as 25% and 19%, respectively of UTI isolates among young females.20,21

In the present study, All strains of S. saprophyticus (100%) were found Nitrofurantoin in contrast with previous studies in which high susceptibility (92%, 100%) of S. saprophyticus against Nitrofurantoin have been reported.<sup>22, 23</sup> However, that study reported S. saprophticus as the most common Gram positive bacteria isolated from UTI among women in accordance with our study.22 Moreover, the 100% sensitivity of S. saprophyticus against imipenem observed in the present study is in consistency with the results of anothe study conducted in Pakistan recently.24

## Conclusion

The study result reveals the antimicrobial sensitivity pattern of S. saprophyticus infections by Imipenem and Amikacin with 100% sensitivity that may help physicians choose the best medications to treat UTIs. Further study requires to explore the mechanism of resistance and control of S. saprophyticus; the most prevalent cause of UTIs in young adult females.

## References

- Argemi X, Hansmann Y, Prola K, Prévost G. Coagulasenegative staphylococci pathogenomics. International journal of molecular sciences. 2019 Mar 11;20(5):1215. https://doi.org/10.3390/ijms20051215
- Pinault L, Chabrière E, Raoult D, Fenollar F. Direct identification of pathogens in urine by use of a specific matrix-assisted laser desorption ionization-time of flight spectrum database. Journal of Clinical Microbiology. 2019 Apr;57(4):10-128.
  - https://doi.org/10.1128/JCM.01678-18
- Govindarajan DK, Kandaswamy K. Virulence factors of uropathogens and their role in host pathogen interactions. The Cell Surface. 2022 Dec 1;8:100075. https://doi.org/10.1016/j.tcsw.2022.100075
- Kline KA, Lewis AL. Gram-positive uropathogens, polymicrobial urinary tract infection, and the emerging microbiota of the urinary tract. Urinary tract infections: Molecular pathogenesis and clinical management. 2017 :459-502.
  - https://doi.org/10.1128/9781555817404.ch19
- Nelson RG, Rosowsky A. Dicyclic and tricyclic diaminopyrimidine derivatives as potent inhibitors of cryptosporidium parvum dihydrofolate reductase: structure-activity and structure-selectivity correlations. Chemotherapy. Antimicrobial Agents and Mar;46(3):940. https://doi.org/10.1128/AAC.02685-20
- 6. K. A. Kline, M. Korte-Berwanger, T. Sakinc, H. V. Nielsen, S. Hultgren, and S. G. Gatermann, "Significance of the d-Serine-deaminase and d-Serine metabolism staphylococcus saprophyticus for virulence," 2013.
- Martins KB, Ferreira AM, Pereira VC, Pinheiro L, Oliveira AD, Cunha MD. In vitro effects of antimicrobial agents on planktonic and biofilm forms of Staphylococcus saprophyticus isolated from patients with urinary tract infections. Frontiers in Microbiology. 2019 28;10:40.https://doi.org/10.3389/fmicb.2019.00040
- A. L. Flores-Mireles, J. N. Walker, M. Caparon, and S. J. J. N. r. m. Hultgren, "Urinary tract infections: epidemiology, mechanisms of infection and treatment options," vol. 13, 5. pp. 269-284. https://doi.org/10.1038/nrmicro3432
- Chua KY, Yang M, Wong L, Knox J, Lee LY. Antimicrobial resistance and its detection in Staphylococcus saprophyticus urinary isolates. Pathology. 2023 Dec

- 1;55(7):1013-6. https://doi.org/10.1016/j.pathol.2023.07.006
- 10. Rabina Dumaru RD, Ratna Baral RB, Shrestha LB. Study of biofilm formation and antibiotic resistance pattern of gram-negative bacilli among the clinical isolates at BPKIHS, Dharan. https://doi.org/10.1186/s13104-019-4084-8
- 11. Spellberg B, Bartlett JG, Gilbert DN. The future of antibiotics and resistance. New England Journal of Medicine. 2013 Jan 24;368(4):299-302. https://doi.org/10.1056/NEJMp1215093
- 12. McQuiston Haslund J, Rosborg Dinesen M, Sternhagen Nielsen AB, Llor C, Bjerrum L. Different recommendations empiric first-choice antibiotic treatment of uncomplicated urinary tract infections in Europe. Scandinavian journal of primary health care. 2013 Dec 1;31(4):235-40.
  - https://doi.org/10.3109/02813432.2013.844410
- 13. Missiakas DM, Schneewind O. Growth and laboratory maintenance of Staphylococcus aureus. Current protocols 2013 microbiology. Feb;28(1):9C-1. https://doi.org/10.1002/9780471729259.mc09c01s28
- 14. Cheesbrough M. District laboratory practice in tropical countries. IJMS. 2018 Jan;1(1):65-8. https://doi.org/10.32441/iims.v1i1.47
- 15. Wayne PA. Clinical and Laboratory Standards Institute: Performance standards for antimicrobial susceptibility testing: 20th informational supplement. CLSI document M100-S20. 2010.
- 16. Ahmed SS, Sharig A, Alsalloom AA, Babikir IH, Alhomoud BN. Uropathogens and their antimicrobial resistance patterns: Relationship with urinary tract infections. International Journal of Health Sciences. 2019 Mar;13(2):48.
- 17. Hayami H, Takahashi S, Ishikawa K, Yasuda M, Yamamoto S, Uehara S, Hamasuna R, Matsumoto T, Minamitani S, Watanabe A, Iwamoto A. Nationwide surveillance of bacterial pathogens from patients with acute uncomplicated cystitis conducted by the Japanese surveillance committee during 2009 and 2010:

- antimicrobial susceptibility of Escherichia coli and Staphylococcus saprophyticus. Journal of Infection and Chemotherapy. 2013 Jan 1;19(3):393-403.
- 18. Kahlmeter GE. An international survey of the antimicrobial susceptibility of pathogens from uncomplicated urinary tract infections: the ECO SENS Project. Journal of antimicrobial Chemotherapy. 2003 Jan 1;51(1):69-76. https://doi.org/10.1093/jac/dkg028
- 19. Marrie TJ, Kwan C. Antimicrobial susceptibility of Staphylococcus saprophyticus and urethral staphylococci. Antimicrobial Agents and Chemotherapy. Sep;22(3):395-7. https://doi.org/10.1128/AAC.22.3.395
- 20. Lo DS, Shieh HH, Barreira ER, Ragazzi SL, Gilio AE. High frequency of Staphylococcus saprophyticus urinary tract infections among female adolescents. The Pediatric infectious disease journal. 2015 Sep 1;34(9):1023-5.https://doi.org/10.1097/INF.0000000000000780
- 21. R. Hague, M. L. Akter, and M. A. J. B. r. n. Salam, "Prevalence and susceptibility of uropathogens: a recent report from a teaching hospital in Bangladesh," vol. 8, pp. 1-5. https://doi.org/10.1186/s13104-015-1408-1
- 22. R. Khoshbakht, A. Salimi, A. H. SHIRZAD, and H. Keshavarzi, "Antibiotic susceptibility of bacterial strains isolated from urinary tract infections in Karaj, Iran," 2013. https://doi.org/10.5812/jjm.4830
- 23. Luty RS, Fadil AG, Najm JM, Abduljabbar HH, Kashmar SA. Uropathogens antibiotic susceptibility as an indicator for the empirical therapy used for urinary tract infections: a retrospective observational study. Iranian Journal of Microbiology. 2020 Oct; 12(5):395. https://doi.org/10.18502/ijm.v12i5.4599
- 24. Khan MA, Rahman AU, Khan B, Al-Mijalli SH, Alswat AS, Amin A, Eid RA, Zaki MS, Butt S, Ahmad J, Fayad E. Antibiotic Resistance Profiling and Phylogenicity of Uropathogenic Bacteria Isolated from Patients with Urinary Tract Infections. Antibiotics. 2023 Oct 3;12(10):1508. https://doi.org/10.3390/antibiotics12101508