ORIGINAL ARTICLE

Prevalence and Antibiotic Sensitivity Pattern of Pseudomonas Aeruginosa Isolates from Urine Samples in A Tertiary Care Hospital

Sohely Sharmin¹, Azizun Nahar², Farhana Alamgir³, Imran Fahim⁴

Abstract

Background: Urinary tract infection (UTI) is one of the leading causes of infection worldwide. P. aeruginosa is a versatile opportunistic pathogen and it is the third most common organism causing nosocomial urinary tract infections. With the widespread use of antibiotics, multidrug resistance pseudomonas continues to go up rapidly. This multidrug resistance pseudomonas increases the risk of mortality and morbidity.

Objective: To determine the prevalence of P. aeruginosa and to understand the current statistics of its antimicrobial resistance pattern.

Methods: This retrospective study was conducted from January 2019 to December 2020 in the Department of Microbiology, Bangladesh Medical College Hospital, Dhaka. Clean catch mid-stream urine samples were collected in sterile containers. The samples were cultured on CLED agar media with a standard calibrated loop and incubated at 37°C overnight. P. aeruginosa grows well at 25°C to 37°C, and its ability to grow at 42°C helped to distinguish it from other Pseudomonas species. Antibiotic sensitivity test was done according to Clinical and Laboratory Standard Institute (CLSI) guideline.

Results: A total of 10427 urine sample were received in the department of Microbiology of BMCH during these two years. Isolation rate of P. aeroginosa was 4% in 2019 and it increased to 6% in 2020. Piperacillin/Tazobactam showed sensitivity which was 63.33% in 2019 and 82% in 2020, followed by imipenem, meropenem, amikacin, ciprofloxacin, ceftazidime and aztreonam; which were 50% to 60% on an average. Sensitivity of the organism to Gentamycin (13.33% in 2019 & 30.3% in 2020), Netilmicin (20% in 2019 & 30.3% in 2020) and Ceftriaxone (13.33% in 2019 & 12.3% in 2020) was found very low.

Conclusion: P. aeruginosa isolates in urine culture is increasing in hospital admitted patient and becoming resistant to multiple antibiotics which is frightening.

Keywords: P. aeruginosa, urine, sensitivity.

- 1. Professor (CC), Department of Microbiology, Bangladesh Medical College.
- 2. Associate Professor, Department of Microbiology, Bangladesh Medical College.
- 3. Professor, Department of Microbiology, Bangladesh Medical College.
- 4. Research Assistant, Bangladesh Medical College, Dhaka.

Correspondence to: Dr. Sohely Sharmin, Professor (CC), Department of Microbiology, Bangladesh Medical College. Cell: 01711955317. E-mail: sohelysharminbmc@gmail.com

Received: 21 August 2021; Accepted: 1 November 2021

Introduction

Urinary tract infection (UTI) is one of the leading causes of infection worldwide. Escherichia coli is predominantly associated with UTI followed by Klebsiella, Proteus, Enterobacter, Citrobacter, Enterococci etc.^{2,3} However non fermenting Gramnegative bacilli such as Pseudomonas is now an emerging pathogen which is an observation of our laboratory and from others.4 Pseudomonas is a ubiquitous, Gram-negative bacillus that can survive in myriad of environment such aquatic and terrestrial.⁵ It is versatile opportunistic pathogen associated with nosocomial infection. The capability of surviving in variety of environmental conditions make it is ubiquitous pathogen, allowing it to persist on numerous living and nonliving surface due to minimum nutritional requirements. According to the report of nosocomial infection surveillance system of center for disease control and prevention P. aeruginosa is the third most common organism causing nosocomial urinary tract infections. Recently this bacterium has acquired resistance to various antibiotics. With the widespread use of antibiotics such as quinolones both in the hospital and community settings, multidrug resistance pseudomonas continues to go up rapidly. This multidrug resistance pseudomonas increases risk of mortality and morbidity. 7-9 Therefore, the aim of the present study was to determine the prevalence of P. aeruginosa in a tertiary care hospital and to understand the current statistics of the antimicrobial resistance pattern of this Gram-negative opportunistic pathogen.

Materials and Methods

The study was conducted in the department of Microbiology, Bangladesh Medical College Hospital (BMCH), Dhaka. This retrospective observational study was conducted over a period of two years from January 2019to December 2020. Samples were received from both inpatient and outpatient department of this hospital. Age and gender of the patients were noted. Clean catch mid-stream urine samples were collected in sterile containers. The samples were cultured on CLED agar (Cystine Lactose Electrolyte Deficient) media with a standard calibrated loop and incubated at 37°C overnight. All the culture and sensitivity reports of urine with positive Pseudomonas aeruginosa showing $\geq 10^5$ colony forming units /ml were considered as significant bacteriuria. The P. aeruginosa isolates were identified by conventional biochemical test. P. aeruginosa grows well at 25°C to 37°C, and its ability to grow at 42°C helps to distinguish it from many other Pseudomonas species. Antibiotic sensitivity test was done by Kirby-Bauer disc diffusion method on Meuller Hinton agar media and interpretation were done according to Clinical and Laboratory Standard Institute (CLSI) guideline. Antibiotics against which susceptibility tested were Ceftriaxone (30μg), Amikacin(30μg), Ciprofloxacin (5μg), ceftazidime(30µg), Cefuroxime (30µg), Imipenem (10µg), Meropenem (10µg), Piperacillin/Tazobactam (110µg), Aztreonam (30µg), Netilmicin (30µg), Gentamycin (10µg) and Colistin (10µg).

Results

A total of 10,427 urine sample were received in the department of Microbiology of BMCH. During these two years out of 10,427, 1337(13%) sample showed growth after culture. Among the positive urine cultures *P. aeroginosa* were identified in 63 (4.7%) cases. Isolation rate of *P. aeroginosa* was 4%in 2019 and it increased to 6% in 2020 (Table I). Out of 63 positive *P. aeroginosa* culture, 51 (81%) was from male. Majority of the samples (84%) were from indoor patient (Table II).

Table I Percentage of Pseudomonas in urine culture						
Year	Total sample	Total growth (%)	Pseudomonas species (%)			
2019	6330	786 (12.42)	30 (3.82)			
2020	4097	551 (13.45)	33 (5.99)			

Table II Distribution of sample according to the age, sex & site of collection (N=65)						
		2019	2020	Total (%)		
Age	Child	2	7	9 (14.29)		
	Adult	28	26	54 (85.71)		
Sex	Male	26	25	51 (80.95)		
	Female	4	8	12 (19.05)		
Site	Indoor	29	24	53 (84.13)		
	Outdoor	1	9	10 (15.87)		

Table III Antibiotic sensitivity pattern of Pseudomonas species						
Drugs	Year					
	2019 N=30	2020 N=33				
Ceftriaxone (CRO)	4 (13.33%)	4 (12.12%)				
Amikacin (AK)	17 (56.66%)	19 (57.57%)				
Ciprofloxacin (CIP)	17 (56.66%)	13 (39.39%)				
Ceftazidime (CAZ)	17 (56.66%)	14 (42.42%)				
Imipenem (IPM)	20 (66.66%)	18 (54.54%)				
Cefuroxime (CXM)	0	1 (3.03%)				
Piperacillin/	19 (63.33%)	27 (81.81%)				
Tazobactum (TZP)						
Meropenem (MEM)	15 (50%)	21 (63.63%)				
Aztreonam (ATM)	16 (53.33%)	14 (42.42%)				
Netilmicin (NET)	6 (20%)	10 (30.30%)				
Gentamicin (GN)	4 (13.33%)	10 (30.30%)				
Colistin (CT)	4 (13.33%)	11 (33.33%)				

Piperacillin/Tazobactam showed higher sensitivity which was 63.33% in 2019 and 82% in 2020, followed by Imipenem, Meropenem, Amikacin, Ciprofloxacin, Ceftazidime and Aztreonam, which were 50% to 60% on an average. Sensitivity of Gentamycin (13.33% in 2019 & 30.3% in 2020), Netilmicin (20% in 2019 & 30.3% in 2020) and Ceftriaxone (13.33% in 2019 & 12.3% in 2020) against P.aeruginosa was found very low (Fig.-1).

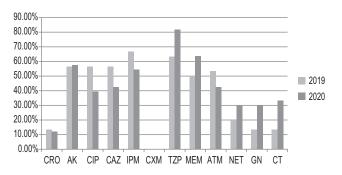


Fig.-1 Antibiotic sensitivity pattern of Pseudomonas species

Discussion

Pseudomonas aeruginosa has become one of the leading causes of hospital acquired as well as community acquired infections due to significant changes in the microbial genetics. Also, the indiscriminate use of antibiotics has resulted rapid spread of acquired multidrug resistance (MDR) that has become global problem.

In our study the rate of isolation of Pseudomonas aeruginosa was 4.7% in urine samples. The isolation rate in the present study is compared to some recent studies from India and other countries. Regha et al 10 from Kerala, India found 3.5% in 2018, Singh et al 3 from Uttar Pradesh, India found 6.75% in 2017 and Shah et al 5 found 5.4% from Karachi in 2015. Even though there is a slight variation in the prevalence, P.~aeruginosa continue to be an important uropathogen in majority of the studies.

When factors such as age, sex in patient and in and outpatient departments were considered, we found that the occurrence of *P. aeruginosa* was higher in male (70%) in adult age. We have also found 85% isolation from indoor patient. Shobha et al¹¹ from Karnataka, India showed 65% male patients in their urine sample had Pseudomonas growth and among them 84% patients were from indoor department. This male preponderance in Pseudomonas infection in urine also goes in concordance with different studies. ^{12,13}

The highest sensitivity of P. aeruginosa isolates was against piperacillin/tazobactam (63% in 2019 and 82% in 2020), imipenem (66.7% in 2019 & 54.5% in 2020), meropenem (50% in 2019 & 63.6% in 2020). Next to this amikacin (56.66% in2019 and 57.57% in 2020). Sensitivity of ciprofloxacin, ceftazidime, aztreonam has been decreased from 2019 to 2020 which is 56.7%

to 39.45%, 56.7% to 42.4%, and 53.3% to 42.4% respectively. In our study imipenem sensitivity decreased from 2019 to 2020 which was 66.7% to 54.5%. Shobha et al^{11,14} found 95.45% Imipenem sensitivity against *P. aeruginosa* in 2015 which reduced to 54.2% in 2017. Regha et al¹⁰ from Kerala, India in 2018 found 72% sensitivity of imipenem which was high in comparison to us. Resistance to carbapenem may be due to the result of complex interaction of several mechanisms including production of carbapenemase, over production of efflux system and loss of outermembrane porins.¹⁵

Present study showed very low sensitivity of gentamycin (13.33% in 2019 & 30.3% in 2020), netilmicin (20% in 2019 & 30.3% in 2020) and ceftriaxone (13.33% in 2019 & 12.3% in 2020) against *P. aeruginosa*. Shobha et al¹¹ found 37.1% sensitivity of gentamycin in 2011 later in 2017 same group found sensitivity 48.6%. ¹⁶ Regha et al¹⁰ found 47% gentamycin sensitivity against *P. aeruginosa* in urine. Pseudomonas resistance to aminoglycosides is probably due to acquisition of resistance genes. Acquisition of aminoglycoside and âlactamresistance gene has been reported in *P. aeruginosa*. ¹⁷⁻¹⁹

Sensitivity of ciprofloxacin has been decreased from 56.7% in 2019 to39.45% in 2020. Principal mode of fluoroquinolone resistance in P. aeruginosa is due to target modifications in DNA gyrase and topoisomerase IV or mutation of regulatory genes of efflux pumps that reduce intracellular concentration of the antibiotic.²⁰

Overall, this low sensitivity of urinary *P. aeruginosa* against commonly used antimicrobials is very alarming. The antibiotic resistance in *P. aeruginosa* is multifactorial in that it can occur through innate, acquired or adaptive mechanisms. The diversity of antibiotics mechanisms contributes to the development of multidrug resistance strains and makes conventional antibiotics ineffective for the treatment of *P. aeruginosa* infections.¹⁵

Conclusion

P. aeruginosa isolates in urine culture is increasing in hospital admitted patient. This organism is becoming resistant to aminoglycoside, fluoroquinolone, carbapenem and β lactam group of antibiotics which is frightening.

References

- Shylaja TS, Mohankumar. A. Antimicrobial Resistance among Uropathogenic Bacteria in Rural Kerala, India. *Int J Curr Microbiol App Sci* 2017; 6:2287-96.
- 2. Tanvir R, Hafeez R, Hasnain S. Prevalence of multiple drug resistant Escherichia coli in patients of urinary tract infection registering at a diagnostic laboratory in Lahore, Pakistan. *Pak J Zool* 2012;44:707-12.
- 3. Singh VP, Mehta A. Bacteriological profile of urinary tract infections at a tertiary care hospital in Western Uttar Pradesh, India. *Int J Res Med Sci* 2017;**5**: 2126-29.
- KL S, Rao GG, Kukkamalla AM. Prevalence of nonfermenters in urinary tract infections in a tertiary care hospital. Webmed Central Microbiology 2011;2(1):WMC001464.doi: 10.9754/ journal. wmc.2011.001464
- Shah DA, Wasim S, Abdullah FE. Antibiotic resistance pattern of Pseudomonas aeruginosa isolated from urine samples of Urinary Tract Infections patients in Karachi, Pakistan. Pak J Med Sci 2015;31:341-45.
- Trivedi MK, Branton A, Trivedi D, Nayak G, Shettigar H, Gangwar M, et al. Antibiogram of Multidrug-Resistant Isolates of Pseudomonas aeruginosa after Biofield Treatment. J Infect Dis Ther 2015;3:244.
- Collee JK, Miles RS. Tests for identification of bacteria. In: Collee JG, Duguid JP, Faser AK, Mermoin BP, editors. Mackie & MacCartney practical Medical Mictobiology. 13th ed.;1989.P141-60.
- 8. Wadud A, Rahman M, Wasey A. Antibiotic resistance in Pseudomonas aeruginosa strains isolated from various clinical specimens. *AFMJ* 2004;**34**:31-35.
- Friedland I, Gallanghor G, King T, Woods GL. Antimicrobial susceptibility pattern in Pseudomonas aeruginosa: data from a multicenter Intensive Care Unit Surveillance Study (ISS) in the US. J Chemother 2004;16:437-41.
- Regha IR. Prevalence and antibiotic susceptibility patterns of pseudomonas aeruginosa in urinary tractSinfections in a Tertiary care hospital, Central Kerala: A retrospective study over 4 rears. Tropical Journal of Pathology & Microbiology 2018:4:52-58.
- Shobha KL, Ramachandra L, Rao AS, Anand KM, Gowsrsh R. PseudomonasSpecies Causing Urinary Tract Infection And Its Antibiogram At A tertiary Care Hospital. Asian J Pharm Clin Res 2017;10: 50-51.

DS (Child) H J 2021; 37 (1)

- Saperston KN, Shapiro DJ, Hersh AL, Copp HL. A comparison of inpatient versus outpatient resistance patterns of pediatric urinary tract infection. *J Urol* 2014;191:5 Suppl:1608-13
- 13. Schaeffer AJ, Nicolle LE. Clinical Practice. Urinary tract infections in older men. N Engl J Med 2016;374:562-71.
- Shobha KL, Souza JS, Alva M, Anand KM, Rao G, Ramachandra L. Study of uropathogens and its antibiogram at a tertiary care hospital at coastal Karnataka. *Ijppr Human* 2015;4:100-09.
- Zheng P, Renee R, Bernard RG, Tong-Jun L, Zhenyu C. Antibiotic resistance in Pseudomonas aeruginosa: mechanisms and alternative therapeutic strategies. *Biotechnology Advances* 2019;37:177-92.
- Shobha KL, Rao GG, Kukkamalla AM. Prevalence of Non-Fermenters in Urinary Tract Infections in a Tertiary Care Hospital. Available from: http:// www.webmedcentral.com.

- 17. de Sa Cavalcanti FL, Mirones CR, Paucar ER, Montes LA, Leal-Balbino TC, de Morais MMC, et al. Mutational and acquired carbapenem resistance mechanisms in multidrug resistant Pseudomonas aeruginosa clinical isolates from Recife, Brazil. *Mem Inst Oswaldo Cruz* 2015;110:1003-1009.
- Hong DJ, Bae IK, Jang IH, Jeong SH, Kang HK, Lee K. Epidemiology and characteristics of Metallo-beta-Lactamase-producing Pseudomonas aeruginosa. *Infect Chemother* 2015;47:81-97.
- 19. Bonomo RA. SzboD. Mechanism of Multidrug Resistance in Acinetobacter species and Pseudomonas aeruginosa. *Clin Infect Dis* 2006;**43**: (suppl2):S49-S56.
- 20. Jalal S, Wretlind B. Mechanisms of quinolone resistance in clinical strains of Pseudomonas aeruginosa. *Microb Drug Resist* 1998;4:257-61.