Section: Microbiology

Original Research Article

EPIDEMIOLOGY AND ANTIMICROBIAL SUSCEPTIBILITY PATTERNS OF CLINICAL PSEUDOMONAS AERUGINOSA AND NON-AERUGINOSA PSEUDOMONAS SPECIES IN A TERTIARY CARE HOSPITAL: A FOUR-YEAR RETROSPECTIVE STUDY

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ABSTRACT

Background: Pseudomonas species are important opportunistic pathogens associated with a wide range of healthcare-associated infections and are recognized for their increasing antimicrobial resistance. Continuous surveillance of resistance patterns are essential for guiding empirical therapy and infection control practices. **Objectives:** To determine the prevalence and antimicrobial susceptibility patterns of clinical Pseudomonas species isolates. **Materials and Methods:** A retrospective observational study was conducted using microbiological records from clinical specimens received over a four-year period at a tertiary care teaching hospital. Pseudomonas isolates recovered from various clinical samples were identified by standard microbiological methods and subjected to antimicrobial susceptibility testing according to institutional laboratory protocols. Demographic characteristics, specimen types, hospital departments, and antimicrobial susceptibility profiles were analyzed. **Results:** Out of 9,000 clinical specimens processed during the study period, 325 (3.6%) yielded Pseudomonas species. The majority of isolates were recovered from pus samples, followed by urine and respiratory specimens. Males accounted for 52% of cases. The highest number of isolates was observed in patients aged 41–50 years. Pseudomonas aeruginosa constituted 34% of all Pseudomonas isolates, while other Pseudomonas species accounted for 66%. Comparatively lower susceptibility was observed for ciprofloxacin and ceftazidime. **Conclusion:** Pseudomonas species continue to be important healthcare-associated pathogens with considerable antimicrobial resistance. Regular surveillance of susceptibility patterns. Continued monitoring of resistance trends is necessary to limit the emergence and spread of multidrug-resistant Pseudomonas isolates.

INTRODUCTION

Pseudomonas species are among the most important opportunistic Gram-negative pathogens causing both community-acquired and healthcare-associated infections. Among them, Pseudomonas aeruginosa is the most clinically significant species and is associated with substantial morbidity and mortality, particularly among hospitalized and immunocompromised patients. Owing to its remarkable metabolic versatility and ability to survive in diverse ecological niches, including hospital environments, P. aeruginosa has emerged as a major cause of nosocomial infections worldwide.[1] Pseudomonas aeruginosa is frequently implicated in pneumonia, urinary tract infections, bloodstream infections, surgical site infections, and ventilator-associated pneumonia. Data from healthcare-associated infection surveillance systems indicate that P. aeruginosa accounts for approximately 7–8% of all healthcare-associated infections and is among the leading causes of nosocomial pneumonia and catheter-associated urinary tract infections.[2] Bloodstream infections caused by P. aeruginosa are particularly concerning because they are associated with mortality rates exceeding 40%, especially in critically ill patients.[2]

The successful persistence of Pseudomonas species in healthcare settings is attributed to their ability to colonize hospital equipment, water sources, disinfectants, respiratory devices, and other moist environments. Their survival on inanimate surfaces

for prolonged periods facilitates transmission and contributes to outbreaks in healthcare facilities.[3] In addition, the organism possesses numerous virulence factors including adhesins, exotoxins, proteases, phenazines, and biofilm-forming capabilities that enhance pathogenicity and promote chronic infection.[4]

Management of Pseudomonas infections is increasingly challenging because of the organism's intrinsic and acquired resistance to multiple classes of antimicrobial agents. Resistance mechanisms include low outer membrane permeability, constitutive expression of multidrug efflux pumps, production of chromosomal AmpC β-lactamases, acquisition of resistance genes through plasmids and transposons, and biofilm formation.[5,6] The coexistence of multiple resistance mechanisms within a single isolate often results in multidrug resistance, thereby limiting therapeutic options and adversely affecting clinical outcomes.[5]

The emergence and dissemination of antimicrobial-resistant Pseudomonas isolates have become a major public health concern. Continuous surveillance of local antimicrobial susceptibility patterns is essential for guiding empirical therapy, informing antimicrobial stewardship programs, and reducing the burden of healthcare-associated infections.[7]

Therefore, the present study was undertaken to determine the prevalence and antimicrobial susceptibility patterns of Pseudomonas species isolated from clinical specimens over a four-year period at a tertiary care hospital in North India.[5-7]

MATERIALS AND METHODS

Study Design and Setting

A retrospective observational study was conducted in the Department of Microbiology, of a tertiary care teaching hospital catering to both urban and rural populations.

Study Period

The study utilized archived microbiology laboratory records from May 2020 to May 2024. All eligible clinical specimens received during the study period were screened for the presence of Pseudomonas species and included in the analysis.

Study Population

Clinical specimens obtained from patients of all age groups and both sexes attending inpatient and outpatient departments of the hospital during the study period were included. Specimens yielding Pseudomonas species on culture were analyzed.

Inclusion Criteria

- Clinical specimens positive for Pseudomonas species.
- Patients of all age groups and both sexes.
- Isolates with complete demographic and antimicrobial susceptibility data.

Exclusion Criteria

- Duplicate isolates obtained from the same patient during the same infectious episode.
- Isolates with incomplete antimicrobial susceptibility records.
- Contaminated specimens or records with incomplete demographic information.

Microbiological Processing and Identification

Clinical specimens including pus, urine, sputum, blood, tissue, and swab samples were processed according to standard microbiological procedures. Specimens were inoculated onto Blood Agar and MacConkey Agar media (HiMedia Laboratories Pvt. Ltd., Mumbai, India) and incubated aerobically at 35–37°C for 18–24 hours.

Identification of Pseudomonas species was performed using conventional microbiological techniques including colony morphology, pigment production, Gram staining, oxidase test, motility testing, and growth at 42°C. Further differentiation of Pseudomonas aeruginosa from other Pseudomonas species was carried out using standard biochemical reactions including oxidative-fermentative glucose utilization, citrate utilization, nitrate reduction, arginine dihydrolase activity, and characteristic pyocyanin and pyoverdine pigment production.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed by the Kirby–Bauer disk diffusion method on Mueller–Hinton Agar (HiMedia Laboratories Pvt. Ltd., Mumbai, India). Zone diameters were interpreted according to the Clinical and Laboratory Standards Institute (CLSI) guidelines applicable during the respective year of testing.

The CLSI standards used during the study period were:

- CLSI M100, 30th Edition (2020)
- CLSI M100, 31st Edition (2021)
- CLSI M100, 32nd Edition (2022)
- CLSI M100, 33rd Edition (2023)
- CLSI M100, 34th Edition (2024)

The antimicrobial disks used (HiMedia Laboratories Pvt. Ltd., Mumbai, India) included:

- Amikacin (30 µg)
- Tobramycin (10 µg)
- Piperacillin–Tazobactam (100/10 µg)
- Ticarcillin–Clavulanic Acid (75/10 µg)
- Ciprofloxacin (5 µg)
- Levofloxacin (5 µg)
- Ceftazidime (30 µg)
- Cefepime (30 µg)
- Aztreonam (30 µg)
- Imipenem (10 µg)
- Meropenem (10 µg)

Quality Control

Quality control for antimicrobial susceptibility testing was performed using Pseudomonas aeruginosa ATCC 27853 in accordance with CLSI recommendations.

Data Collection and Statistical Analysis

Data regarding patient demographics, specimen type, hospital department, bacterial identification, and antimicrobial susceptibility results were extracted

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from laboratory records and entered into Microsoft Excel. Data were summarized using frequencies and percentages.

Ethical Considerations

The study was approved by the Institutional Ethics Committee of Integral Institute of Medical Sciences and Research, Lucknow (Approval No. IEC/IIMSR/2024/65). As this was a retrospective record-based study utilizing anonymized laboratory data, patient confidentiality.

RESULTS

During the study period (May 2020–May 2024), 9,000 clinical specimens were received and processed. Among these, 325 (3.6%) yielded Pseudomonas species and were included in the final analysis.

Table 1: Demographic and Clinical Characteristics of Patients with Pseudomonas Species Isolates (n = 325)

Variable	Frequency (n)	Percentage (%)
Gender		
Male	170	52.3
Female	155	47.7
Age Group (years)		
0–10	11	3.4
11–20	55	16.9
21–30	45	13.8
31–40	67	20.6
41–50	81	24.9
51–60	38	11.7
>60	28	8.6
Clinical Specimen		
Pus	160	49.2
Urine	96	29.5
Sputum	47	14.5
Blood	16	4.9
Swab	5	1.5
Tissue	1	0.3
Hospital Department		
Medicine	111	34.2
ENT	66	20.3
Surgery	50	15.4
Orthopedics	42	12.9
Emergency	29	8.9
ICU	17	5.2
Obstetrics & Gynecology	10	3.1
Species Distribution		
Pseudomonas aeruginosa	109	33.5
Other Pseudomonas species	216	66.5

Table 2: Antimicrobial susceptibility pattern of Pseudomonas species isolates (n = 325)

Antibiotic	Sensitive (n)	Resistant (n)	Total Tested (n)	Susceptibility (%)	Resistance (%)
Amikacin (AK)	162	41	203	79.8	20.2
Tobramycin (TOB)	154	56	210	73.3	26.7
Piperacillin-Tazobactam (PIT)	155	51	206	75.2	24.8
Ticarcillin-Clavulanate (TCC)	159	47	206	77.2	22.8
Ciprofloxacin (CIP)	136	69	205	66.3	33.7
Levofloxacin (LEVO)	149	56	205	72.7	27.3
Ceftazidime (CAZ)	142	63	205	69.3	30.7
Cefepime (CPM)	165	51	216	76.4	23.6
Aztreonam (AT)	149	53	202	73.8	26.2
Imipenem (IPM)	174	51	225	77.3	22.7
Meropenem (MRP)	152	56	208	73.1	26.9
Norfloxacin (NX)	103	29	132	78	22

Abbreviations: AK, amikacin; TOB, tobramycin; PIT, piperacillin-tazobactam; TCC, ticarcillin-clavulanate; CIP, ciprofloxacin; LEVO, levofloxacin; CAZ, ceftazidime; CPM, cefepime; AT, aztreonam; IPM, imipenem; MRP, meropenem; NX, norfloxacin.

Of the 325 isolates, 170 (52.3%) were recovered from male patients and 155 (47.7%) from female patients, yielding a male-to-female ratio of 1.1:1.

The highest proportion of isolates was observed in patients aged 41–50 years (81/325; 24.9%), followed by 31–40 years (67/325; 20.6%) and 11–20 years (55/325; 16.9%). The lowest frequency was observed in children aged 0–10 years (11/325; 3.4%).

Distribution of Isolates by Clinical Specimen

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Pus was the most common specimen yielding Pseudomonas isolates, accounting for 160 (49.2%) isolates, followed by urine [96 (29.5%)], sputum [47 (14.5%)], blood [16 (4.9%)], swab [5 (1.5%)], and tissue samples [1 (0.3%)].

Distribution of Isolates by Hospital Department

The highest number of Pseudomonas isolates was obtained from the Department of Medicine [111 (34.2%)], followed by ENT [66 (20.3%)], Surgery [50 (15.4%)], Orthopedics [42 (12.9%)], Emergency [29 (8.9%)], Intensive Care Unit [17 (5.2%)], and Obstetrics and Gynecology [10 (3.1%)].

Species Distribution

Among the 325 isolates, Pseudomonas aeruginosa accounted for 109 (33.5%) isolates, whereas other Pseudomonas species constituted 216 (66.5%) isolates.

Antimicrobial Susceptibility Pattern table 2

Antimicrobial susceptibility testing revealed variable susceptibility rates across the tested antibiotics. The highest susceptibility was observed for amikacin [241/306; 78.8%], cefepime [255/325; 78.5%], aztreonam [241/309; 78.0%], and ticarcillin–clavulanic acid [240/309; 77.7%].

Piperacillin–tazobactam demonstrated a susceptibility rate of 77.5% (241/311), followed by levofloxacin [231/305; 75.7%], imipenem [227/300; 75.7%], tobramycin [238/317; 75.1%], and meropenem [237/316; 75.0%].

Comparatively lower susceptibility was observed for ceftazidime [226/309; 73.1%] and ciprofloxacin [212/297; 71.4%], indicating relatively higher resistance to fluoroquinolones and third-generation cephalosporins.

Overall, resistance rates ranged from 21.2% for amikacin to 28.6% for ciprofloxacin among the tested isolates.

DISCUSSION

Pseudomonas species are important opportunistic pathogens associated with a wide range of healthcare-associated infections and are increasingly recognized as major contributors to antimicrobial resistance worldwide. Their remarkable ability to survive in hospital environments, form biofilms, and acquire multiple resistance mechanisms has made them a persistent challenge for clinicians and microbiologists alike.[1,5,6]

In the present study, Pseudomonas species were isolated from a diverse range of clinical specimens, with pus being the most common source (49.2%), followed by urine (29.5%) and sputum (14.5%). Similar findings have been reported by Javiya et al,[8] who observed wound and pus specimens as the predominant source of Pseudomonas isolates in a tertiary care setting. The high recovery rate from pus samples may be attributed to the organism's ability to colonize chronic wounds, postoperative sites, and burn injuries, where biofilm formation contributes to persistent infection and delayed healing.[4,5]

A slight male predominance (52.3%) was observed in the present study, which is comparable to findings reported by Senthamarai et al,[9] and Aggarwal et al.[10] The higher frequency among males may be related to greater occupational exposure, increased hospitalization rates, and a higher prevalence of trauma-related infections. The majority of isolates were recovered from patients aged 41–50 years, suggesting that middle-aged adults constitute an important risk group for Pseudomonas infections. Similar age distributions have been reported in previous Indian studies.[9,10]

Among the isolates recovered, Pseudomonas aeruginosa accounted for 33.5% of all Pseudomonas species. Although non-aeruginosa Pseudomonas species constituted a larger proportion of isolates, P. aeruginosa remains the most clinically significant member of the genus because of its established role in severe healthcare-associated infections and multidrug resistance.[1,2]

The antimicrobial susceptibility profile demonstrated that amikacin exhibited the highest (79.8%). These findings are consistent with reports by Goli et al,[11] and Pathmanathan et al,[12] who observed high susceptibility of Pseudomonas isolates to polymyxins and aminoglycosides. The preserved activity of these agents may be due to their restricted clinical use compared with other antibiotic classes. However, the emergence of resistance even to reserve drugs warrants continuous surveillance and judicious antimicrobial use.

Among β-lactam agents, piperacillin–tazobactam, ticarcillin–clavulanate, cefepime, and aztreonam demonstrated susceptibility rates exceeding 75%. Similar susceptibility patterns have been reported from tertiary care centers across India.[13,14] These findings suggest that β-lactam/β-lactamase inhibitor combinations continue to retain useful activity against a substantial proportion of Pseudomonas isolates in our setting.

Ciprofloxacin demonstrated the lowest susceptibility rate (66.3%) and the highest resistance rate (33.7%) among the antibiotics tested. Increasing resistance to fluoroquinolones has been reported globally and is largely attributed to widespread empirical use, target-site mutations, and overexpression of efflux pumps.[5,6] Comparable ciprofloxacin resistance rates have been documented by Sharma et al,[15] and Sivanmaliappan and Sevanan,[16] emphasizing the declining utility of fluoroquinolones for empirical therapy against Pseudomonas infections.

Carbapenems remain among the most effective therapeutic options for serious Pseudomonas infections. In the present study, susceptibility rates for imipenem, meropenem, were 77.3%, 73.1%, respectively. Although these rates remain relatively favorable, the observed resistance highlights the growing concern regarding carbapenem-resistant Pseudomonas species. The development of carbapenem resistance is often mediated through porin loss, efflux pump overexpression, AmpC hyperproduction, and carbapenemase production.[5,6]

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Similar resistance trends have been reported in studies from other tertiary care hospitals in India.[13,15]

These findings suggest that resistance patterns may vary according to patient demographics and bacterial species and underscore the importance of local surveillance data for guiding empirical antimicrobial therapy.[14,17]

From an infection prevention and control perspective, the predominance of Pseudomonas isolates in pus, urine, and respiratory specimens highlights the need for strict adherence to standard infection control practices, including hand hygiene, environmental cleaning, device-care bundles, and surveillance of healthcare-associated infections. Pseudomonas species are known to persist in moist hospital environments, medical equipment, sinks, water outlets, and respiratory devices, facilitating nosocomial transmission and outbreaks. Therefore, periodic microbiological surveillance and prompt implementation of infection control interventions are essential to prevent the dissemination of resistant strains within healthcare facilities.[2,3]

The findings of the present study also have important implications for antimicrobial stewardship programs (AMSP). The relatively high resistance observed against ciprofloxacin and moderate resistance to carbapenems emphasize the need for judicious antimicrobial use and regular review of empirical antibiotic policies. Institutional antibiograms should be updated periodically and integrated into stewardship activities to guide evidence-based empirical therapy. Preservation of the efficacy of reserve agents such as polymyxins and carbapenems should remain a priority, and unnecessary use of broad-spectrum antimicrobials should be discouraged. Continuous collaboration between microbiologists, infectious disease specialists, clinicians, pharmacists, and infection control teams is essential to optimize antimicrobial utilization and mitigate the emergence of multidrug-resistant Pseudomonas isolates.[5,6,17]

CONCLUSION

The findings underscore the importance of continuous antimicrobial resistance surveillance, robust infection prevention and control practices, and implementation of effective antimicrobial stewardship programs to limit the emergence and spread of resistant Pseudomonas species in healthcare settings.

Limitation

Limitations of the study include its retrospective design, reliance on laboratory records, and the absence of molecular characterization of resistance mechanisms. Future prospective studies incorporating molecular resistance profiling and multidrug-resistance classification would provide a more comprehensive understanding of the epidemiology of Pseudomonas infections.

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