Original Article

Antimicrobial Resistance Dynamics of *Pseudomonas Aeruginosa* in a Tertiary Care Hospital in Delhi

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Abstract

Background: *Pseudomonas aeruginosa*, non-fermenting gram-negative bacilli are a major cause of healthcare-associated infections. The rise in multidrug-resistant (MDR) strains globally has been associated with increased morbidity and mortality. In India, antimicrobial resistance (AMR) presents a significant challenge, necessitating effective policies and infection control measures. This study explores the resistance dynamics of *P. aeruginosa* in a tertiary care hospital, evaluating the findings in relation to the data from the Indian Council of Medical Research (ICMR).

Methods: A total of 1,536 *P. aeruginosa* isolates from 31997 clinical samples of pus, wound, and burn swabs, respiratory specimens and body fluids submitted for routine culture were analyzed. Antimicrobial susceptibility testing was performed using the Kirby-Bauer disk diffusion method. Blood, urine, cerebrospinal fluid (CSF), and stool were excluded.

Results: *P. aeruginosa* accounted for 5th most frequent pathogen with prevalence declining from 15.7% (136/869) in 2020 to 7.7% (249/3236) in 2024. Among the isolates, 34.2% (525/1536) were susceptible to all tested antimicrobials, while 13.2% (203/1536) were MDR and 10.6% (163/1536) were extensively drug-resistant (XDR). Improved susceptibility was observed for piperacillin-tazobactam, carbapenems, aztreonam, and aminoglycosides, whereas, fluoroquinolones and monobactams showed persistently low susceptibility.

Conclusion: Continuous monitoring of MDR pathogens like *P. aeruginosa* and their resistance dynamics are crucial for guiding treatment strategies and infection control measures.

Keywords: pseudomonas aeruginosa; antimicrobial resistance; hospital-acquired infections; icmr

Introduction

Pseudomonas aeruginosa, a non-fastidious, non-fermenting gram-negative bacilli is ubiquitously found in nature. The pathogen is leading cause of healthcare-associated infections (HAI), estimated to cause 10-11% of all HAIs, including pulmonary infections (ventilator-associated pneumonia), surgical site infections, urinary tract infections (UTI) particularly catheter-associated, infection in burn-wound patients, and is often associated with high morbidity and mortality [1].

Due to its intrinsic resistance to many antimicrobials, anti-*pseudomonal* aminoglycosides, β-lactam/β-lactamases inhibitors (BL-BLI), anti-*pseudomonal* cephalosporins, and fluoroquinolones are first-line treatments for mild to moderate *Pseudomonal* infections [2]. Aminoglycosides exhibit broad-spectrum, concentration-dependent killing by irreversibly binding to the

30S ribosomal subunit, inhibiting bacterial protein synthesis. However, monotherapy is not recommended for systemic infections (except UTIs); combination therapy is preferred [3]. Piperacillin–tazobactam, a BL-BLI with activity against gram-positive, gram-negative, and anaerobic bacteria. It remains effective against broad-spectrum and some extended-spectrum β-lactamase-producing (ESBL) bacteria, but is inactive against AmpC β-lactamase-producing gram-negative bacilli. It is compatible with gentamicin and amikacin but not tobramycin. With its excellent safety profile, piperacillin–tazobactam remains a reliable choice for empiric treatment of moderate-to-severe *Pseudomonal* infections [4]. Ceftazidime, a third-generation cephalosporin with broad-spectrum activity, including against *P. aeruginosa*, is effective for lower respiratory and complicated UTI in hospitalized patients. It is resistant to β-lactamases and offers a safer alternative to aminoglycosides, with minimal toxicity and does not necessitate drug plasma concentration monitoring [2]. Fluoroquinolones constitute up to 25% of antibiotic use in India. Ciprofloxacin, the most prescribed fluoroquinolone, targets DNA (Deoxyribonucleic Acid) gyrase and topoisomerase IV, essential for bacterial DNA synthesis, and is effective against non-fermenters like *Pseudomonas* [5].

Overuse of antibiotics exerts selective pressure on microorganisms, leading to the development of resistant strains. *P. aeruginosa* is particularly challenging due to its ability of acquiring resistance during antibiotic therapy, resistance to antiseptics, and disinfectants leading to higher mortality, costs and prolonged hospital stays [6]. Centers for Disease Control and Prevention (CDC) reported that 13% of *P. aeruginosa* HAIs are caused by multidrug-resistant (MDR) strains, which are resistant to at least one antibiotic in three classes of antibiotics [7, 8]. Carbapenems, broad-spectrum β-lactam antibiotics with time-dependent bactericidal action, are the treatment of choice for MDR *Pseudomonal* infections. However, resistance mechanisms such as carbapenemases production, porin loss, or efflux pumps, have led to carbapenem-resistant strains [8, 9]. Several studies highlight the emergence of extensively drug-resistant *P. aeruginosa* (XDR-PA), defined as insusceptibility of isolate to at least one antibiotic in all, but two or more antimicrobial categories. The resistance is further amplified due to colistin-resistance enzymes, by integron-associated resistance genes and biofilm formation in addition to other β-lactamases production [7, 8, 9]. The World Health Organization (WHO) has classified carbapenem-resistant *P. aeruginosa* (CRPA) as high priority pathogen for research and development of new antimicrobials. CRPA are often untreatable, resulting in high treatment failure rates, also associated with nosocomial spread [10].

Aztreonam, a reserve monobactam as per WHO (Access, watch & Reserve classification) AWaRe, is effective against Class B Metallo-β-lactamases (MBLs), which confer resistance to most β-lactams, including carbapenems. However, its efficacy is limited in MDR/XDR strains which co-produce MBLs with ESBLs or AmpC, restricting treatment options [11]. To overcome this, aztreonam can be combined with advanced β-lactamase inhibitors like avibactam, zidebactam, nacubactam, or taniborbactam, offering new therapeutic possibilities [11]. However, apart from aztreonam/avibactam, these combinations are not yet accessible in low-resource settings with heavy patient volumes as seen in India. In such scenarios, reliance on available antimicrobials, knowledge of local antimicrobial resistance (AMR) data and stringent hospital infection control practices remains the primary strategy to curb the transmission of MDR /XDR *Pseudomonas* strains. Studies indicate that contact precautions in Intensive Care Units (ICUs) can reduce MDR *P. aeruginosa* infection rates, while standard precautions alone do not increase hospital infection rates [6]. India faces AMR and the challenge of implementing effective policies and infection control measures as significant health challenges. The Government of India launched National Action Plan on Antimicrobial Resistance (NAP-AMR) 2017–2021 and the Indian Council of Medical Research (ICMR) established the Antimicrobial Resistance Surveillance and Research Network (AMRSN) to comprehend the patterns and scope of the issue [12, 13]. Understanding the dynamics of prevalent MDR/XDR strains and antimicrobial susceptibility of *P. aeruginosa* in local settings is crucial for adopting pivotal infection strategies [6]. This study explores the resistance dynamics of *P. aeruginosa* in a tertiary care hospital setting evaluating the data alongside ICMR.

Material & Methods

Study area and duration

A retrospective observational study was conducted over a period of four and a half years (January 2020 to July 2024) in the bacteriology laboratory of Department of Microbiology, University College of Medical Sciences & associated Guru Teg Bahadur (GTB) hospital, Delhi.

Samples size

A total of 1536 non-duplicate isolates of *P. aeruginosa* isolated from 31977 various clinical samples including samples of pus, wound swabs, respiratory tract, body fluids, burn wounds and other samples (ear swabs, tissue biopsy etc.), which were received in the bacteriology lab for routine culture and antimicrobial susceptibility testing (AST) were included in the study. Samples of Blood, Urine, and CSF were excluded from the study.

Method

The samples were processed conventionally as per standard laboratory protocol. The samples were streaked on Blood agar and MacConkey agar media for isolation of the causative pathogen, followed by identification of the bacterial isolates by series of biochemical test. The resistance profiles of identified *P. aeruginosa* for various classes of antimicrobials including Aminoglycosides (Amikacin 30μg, Gentamicin 10μg and Tobramycin 10μg), BL-BLIs (Piperacillin-tazobactam 100/10μg), Third generation Cephalosporins (Ceftazidime 30μg), Fluoroquinolones (Ciprofloxacin 5μg), Carbapenems (Imipenem 10μg and Meropenem 10μg) and Monobactams (Aztreonam 30μg) were tested by Kirby-Bauer disc diffusion method. An inoculum equivalent to 0.5 McFarland standard was lawned onto Mueller-Hinton agar and the antibiotic disks (procured from HiMedia laboratories, India) were placed. The plates were then incubated at 35° C aerobically for 16-18 hours.

The isolates which were resistant to at least one antibiotic in three classes of antibiotics (Aminoglycosides, BL-BLIs, Third generation Cephalosporins, Fluoroquinolones, Carbapenems and Monobactams) were considered MDR, and isolates which were resistant to at least one antibiotic in all, but two or more antimicrobial classes were considered as XDR [14].

For quality control, *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* (ATCC 25923) and *Pseudomonas aeruginosa* (ATCC 27853) strains were tested daily by Kirby-Bauer disc diffusion method as a part of routine laboratory practice. The zone of inhibitions were interpreted according to the CLSI guidelines of the respective year, with results compared to the acceptable QC ranges for each antibiotic [15, 16, 17, 18, 19].

Interpretation

The results were interpreted as per the Clinical and Laboratory Standards Institute (CLSI-M100) guidelines of the year [15, 16, 17, 18, 19].

Data analysis

The data analysis was done by using World Health Organization Network (WHONET) software. The result was expressed as percentages and graphs were generated with the help of WHONET and Microsoft Excel software. Yearly prevalence and antimicrobial susceptibility trends of *P. aeruginosa* were evaluated using the Pearson's Chi-square test. Differences in proportions across years were assessed using contingency tables, with p value < 0.05 considered statistically significant.

Results

Prevalence dynamics of *Pseudomonas aeruginosa*

During the study period, 31,977 clinical specimens of pus, body fluids, and respiratory tract specimens, were submitted for routine culture and sensitivity testing, yielding 15,503 (48.4%) bacterial isolates. The yearly bacterial culture positivity varied between 46.2% and 53.3% during the study period (Figure 1). The overall prevalence of *P. aeruginosa* was 10%

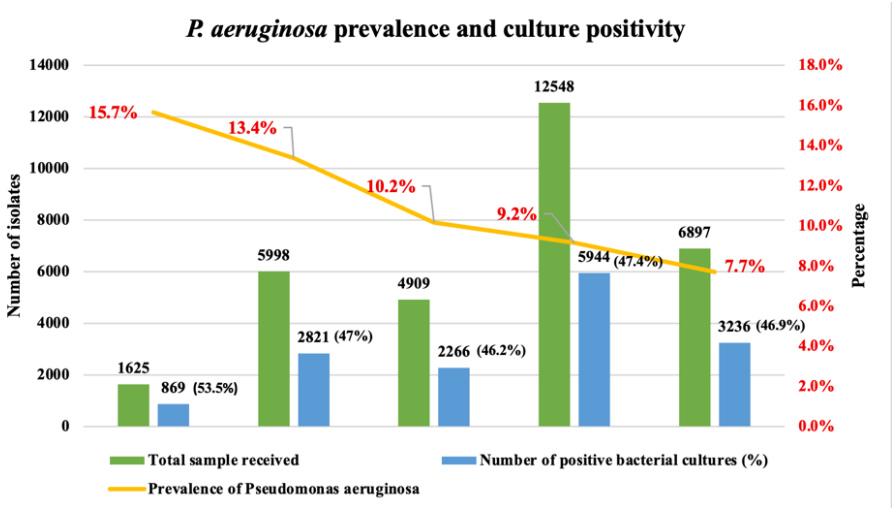

Figure 1: Prevalence of *P. aeruginosa* and culture positivity (p value < 0.00001).

(1536/15503) (Figure 2). The prevalence of *P. aeruginosa* declined from 15.7% (136/869) in 2020 to 7.7% (249/3236) till

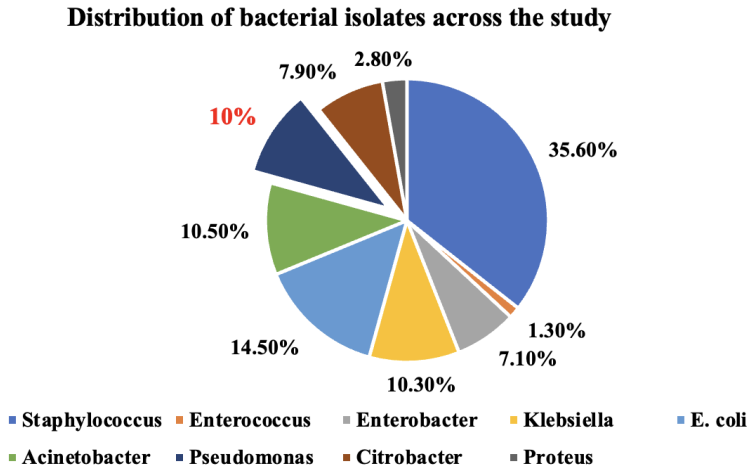

Figure 2: Distribution of bacterial isolates across the study.

June 2024. Year-wise prevalence was 13.4% (378 / 2821) in 2021, 10.2% (231 / 2266) in 2022, and 9.2% (546 / 5944) in 2023. This decline was statistically significant ($\chi^2 = 95.75$, $p < 0.00001$). The *P. aeruginosa* were predominantly isolated from the samples of pus 82% (1264), wound swabs 8.4% (130), respiratory tract 5.4% (83), body fluids 1.3% (21), burn wounds 1%(14) and 1.5% (24) from other samples (ear swabs, tissue biopsy etc.) Among the isolated *P. aeruginosa*, 40% (614) were obtained from out-patient department (OPD), 56% (860) from inpatient department (IPD), and 4% (62) from ICU. Of the total *P. aeruginosa* isolates, 59% (906) were from male patients and 41% (630) from females. Patient demographics were limited to sample type, patient location (OPD/IPD/ICU), and gender, as no additional clinical or demographic information was collected.

Distribution of common bacterial pathogens

Out of the 15503 isolated bacteria, 1536 (10%) were *P. aeruginosa*, accounted for 5th most frequent pathogen after *Staphylococcus aureus*, *Escherichia coli*, *Acinetobacter baumannii* complex and *Klebsiella pneumoniae*. The overall distribution of common pathogens and their annual isolation trends are given in the figure 2 and 3 respectively.

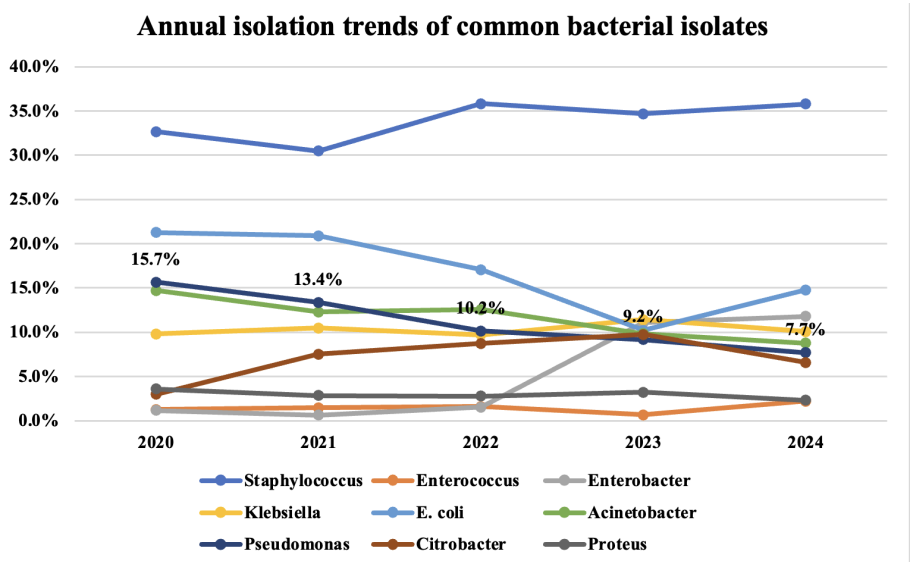

Figure 3: The annual isolation trends of common bacterial isolates.

Susceptibility dynamics of *Pseudomonas aeruginosa*

Among 1536 isolated *P. aeruginosa*, an average of 34.2% (525/1536) of isolates were susceptible to all the tested antimicrobial across the study. However, 13.2% (203/1536) of isolates were MDR, and 10.6% (163/1536) were XDR (Supplementary

image 1 and 2). The annual susceptibility trend of *P. aeruginosa* (Figure 4) showed significant variation over time (χ^2 = 45.31, p < 0.00001).

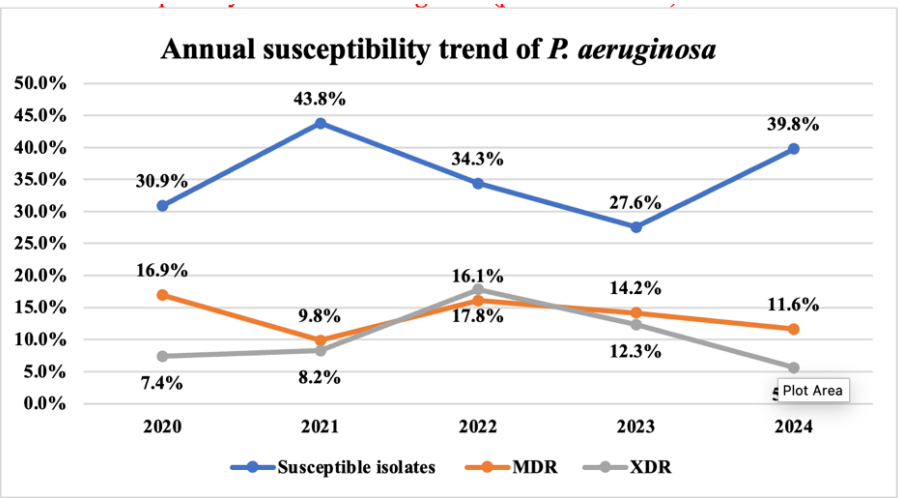

Figure 4: Annual susceptibility trends of *P. aeruginosa* (p value < 0.00001). MDR: Multi-drug resistant; XDR: Extensive drug resistance.

The antimicrobial susceptibility to various antimicrobials

Figure 5 presents the antimicrobial susceptibility profile of *P. aeruginosa* to various antimicrobial classes throughout the study. The figure 5 also includes susceptibility data for *P. aeruginosa* (of 11,757 isolates from all samples other than feces) from the ICMR Antimicrobial Resistance Surveillance Network's annual report (January to December 2023) [20].

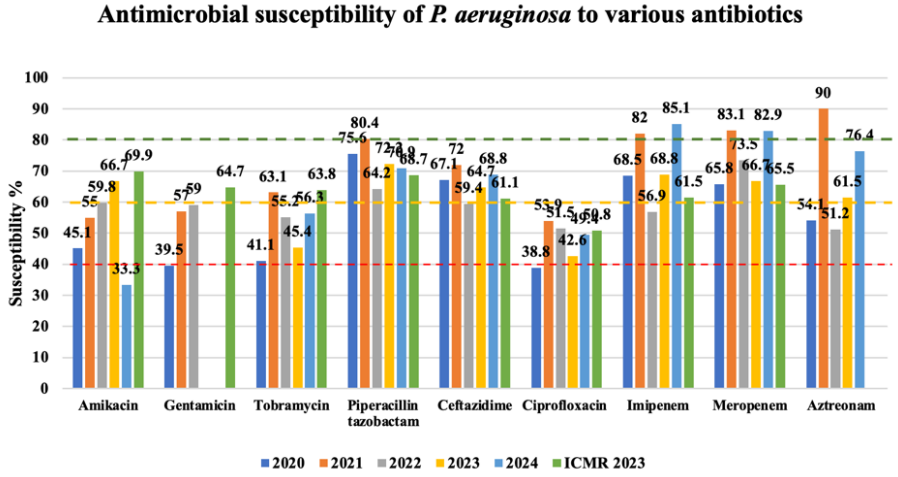

Figure 5: The trends in antimicrobial susceptibility of *P. aeruginosa* to various antibiotics. ICMR: Indian Council of Medical Research.

Discussion

P. aeruginosa is a major healthcare-associated pathogen, responsible for up to 11% of all HAIs, causing various disease manifestations, comprising 10% of the isolates identified in our study highlighting its significant local burden [1]. The prevalence significantly reduced to almost half over the last 4 and half year (15.7% to 7.7%). The decline was nearly twice in the first two years, likely due to stringent hospital infection control measures implemented during the COVID-19 pandemic, compared to the subsequent years. As per the ICMR data, the yearly isolation trend of *Pseudomonas* isolates from all the samples reduced from 15% in 2016 to 11.8% 2023 [20, 21]. This consistent declining trend of *P. aeruginosa* prevalence from our study aligns with that of ICMR. In this study, we observed a higher prevalence of *P. aeruginosa* infections in males (59%) compared to females (41%), consistent with findings by Vincent et al. [22]. In our study, most *P. aeruginosa* isolates were from inpatients (56%), followed by outpatients (40%). Only 4% of isolates were from ICU samples. In contrast, Vincent et al. reported that *P. aeruginosa* accounted for 16.2% of infections in ICU patients [22]. Similarly, Harris et al.

found that 11.6% of ICU patients were colonized with *P. aeruginosa* at admission, making them over six times more likely to develop infections caused by this pathogen than non-colonized patients [22, 23]. The lower prevalence of *P. aeruginosa* in the ICU in our study may be attributed to the exclusion of blood and urine samples, which were predominant sources of *P. aeruginosa* in these studies. These observations highlight key areas for local infection control measures, including targeted monitoring of high-risk patient populations, adherence to bundle care in surgical site infections, reinforcement of hygiene and wound management practices, and prioritization of preventive measures in inpatient settings.

The emergence of AMR to *P. aeruginosa* poses a significant public health challenge, complicating treatment options. The CDC identifies MDR *P. aeruginosa* as a serious threat and one of seven high-burden antimicrobial-resistant pathogens in healthcare settings [24]. The INFORM (International Network for Optimal Resistance Monitoring) database reported the rates of MDR *P. aeruginosa* ranged from 11.5% to 24.7% of total isolates [25]. In this study, overall, 34.2% *P. aeruginosa* isolates, were susceptible to all the tested antimicrobial while 13.2% were MDR, and 10.6% were XDR. In 2021, the prevalence of susceptible isolates was higher, and MDR was lower compared to other years. This could be due to robust infection control practices due to COVID-19 pandemic. However, MDR prevalence remained above 11% and XDR above 5% in the remaining years, highlighting that need for continued infection control measures is crucial to sustain the reduction in MDR *P. aeruginosa* infections. Al-Orphaly et al. reported a high prevalence of MDR *P. aeruginosa* in Egypt (75.6%), moderate prevalence in Saudi Arabia (7.3%) and Qatar (8.1%) comparable to our study, while the rates were lowest in Morocco (0%) [26]. Similarly, Ullah et al. reported that over 30% of *P. aeruginosa* isolates in acute exacerbation of Chronic Obstructive Pulmonary Disease (COPD) cases in South India were XDR before 2016. However, from 2016 to 2020, XDR rates declined while MDR rates increased [27].

Aminoglycosides, anti-*pseudomonal* cephalosporins, BL-BLIs and fluoroquinolones are commonly used antimicrobials for *P. aeruginosa* infections. In critically ill patients or those with known colonization or risk factors for MDR infections, reserved antibiotics, including carbapenems, novel BL-BLIs, monobactam, and polymyxins, are often indicated [9]. In our study, the susceptibility to aminoglycosides improved from approximately 40% to 65% over time. These findings align with ICMR data and may be attributed to the limited use of aminoglycosides as mono-therapy in infections other than uncomplicated UTIs [19, 20]. The trend was more consistent with amikacin compared to tobramycin. In 2023, routine testing for amikacin was reduced as it is primarily recommended for urinary isolates. Conversely, tobramycin, remains a first line treatment option alongside piperacillin-tazobactam for mild to moderate *Pseudomonas* infections, which may have contributed to the observed inconsistency in the tobramycin susceptibility trend. The gentamicin testing was discontinued in 2023 and 2024 due to non-availability of breakpoints in CLSI guidelines of those years [19]. Saeli et al. reported resistance rates of 45.5% to tobramycin and 43% to amikacin in clinical strains of 200 *P. aeruginosa*, aligning with findings from our study [28].

Ceftazidime, a tier 1 antimicrobial as per CLSI, has strong activity against non-fermenters including *Pseudomonas* [19]. In our study, susceptibility to ceftazidime ranged from 60–70%, aligning with ICMR data [20]. Similar to our study, Bazghandi et al. reported 53.6% susceptibility to ceftazidime [7]. In contrast, Krovvidi et al. observed 68.6% ceftazidime resistance in *P. aeruginosa* from various samples while Saeli et al. reported high resistance rate of 86.4% in MDR *P. aeruginosa* [28, 29]. Ceftazidime is easily hydrolyzed by ESBLs, AmpC, and carbapenemases enzymes. Thus, BL-BLI combinations or ceftazidime-avibactam should be considered to enhance activity against ESBL, AmpC, and *Klebsiella pneumoniae* carbapenemase (KPC) producers.

Among BL-BLI combinations, piperacillin-tazobactam is one of the most commonly prescribed antibiotics in critically ill patients with moderate to severe infections [2, 4]. In this study, piperacillin-tazobactam exhibited approximately 70% susceptibility across *Pseudomonal* isolates, with minimal variation in susceptibility observed each year. These findings are consistent with the data reported by ICMR [20]. Al-Orphaly et al. reported resistance rates of piperacillin-tazobactam in *Pseudomonas* ranging from 7% to 42.3% in Middle East and North Africa region, aligning with our findings [26]. However, the susceptibility observed in our study was higher compared to Bazghandi et al. and Reddy et al. who reported 53.6% and 31% susceptibility to piperacillin-tazobactam in *Pseudomonas* isolated from ICU settings respectively [7, 30]. These differences may be attributed to variations in their sample types and the exclusive inclusion of ICU patients. As per WHO, in 2022, fluoroquinolones had the highest resistance percentage (19.6%) among antibiotics in the EU/EEA (European Union and the European Economic Area), followed by piperacillin-tazobactam (18.8%) [31]. In this study, ciprofloxacin susceptibility remained consistently low at 40-50% throughout the study period, aligning with ICMR data [20]. Similar findings have been documented in other studies, such as Reddy et al. reported 41% susceptibility, and Krovvidi et al. reported 45-48% susceptibility to fluoroquinolones [29, 30]. However, contrasting results were noted by Lastinger et al. who reported lower resistance (27.1%) among *Pseudomonas* isolates in ICU settings, whereas Al-Orphaly et al. observed a significantly higher resistance rate (72-100%) among multidrug-resistant isolates [26, 32]. Fluoroquinolones are known for their broad-spectrum activity, excellent oral absorption, and high tissue penetration, particularly in the respiratory system due to their large volume of distribution. Their oral availability makes them a common choice for outpatient treatment, especially in patients with chronic lung diseases like COPD with mild to moderate severity [5]. This frequent use in such settings could contribute to the observed variability in susceptibility across different studies.

Carbapenems, a superior class of β-lactam antibiotics, is stable to β-lactamase enzymes and serves as the treatment of choice

for MDR *Pseudomonal* infections. However, carbapenem-resistant or presence of carbapenemases producing *P. aeruginosa* remains a significant healthcare challenge globally due to increasing probability of treatment failures [10]. As per WHO data from 2020, in European regions, 10% of countries reported CRPA rates below 5%, while 15% had rates of 50% or higher, with the remainder falling between 5% and 50% [31]. In contrast, in the United States, 10–30% of *P. aeruginosa* isolates were carbapenem-resistant [33]. In our study, susceptibility to imipenem and meropenem ranged from 57% to 85%. Notably, the prevalence of MDR and XDR pathogen was lower in 2021 and 2024. During these years, susceptibility rates to imipenem and meropenem were higher, exceeding 80%, while less favourable trends were observed over the remaining study period. Overall, imipenem susceptibility in our study was slightly higher than ICMR data, while meropenem susceptibility was comparable [20]. Other studies have reported varying susceptibility patterns to carbapenems. Bazghandi et al. observed lower susceptibility to imipenem (34%) and comparable susceptibility to meropenem (57%) [7]. Krovvidi et al. reported resistance rates of 45% for imipenem and 50% for meropenem in *P. aeruginosa* isolates from various samples [29]. Similarly, Varaiya et al. documented 71% susceptibility to carbapenems among respiratory isolates from ICU patients, while Reddy et al. reported 40% susceptibility from ICU samples [30, 34]. The variation in carbapenem susceptibility may stem from gaps in post-COVID surveillance and infection control practices. Additionally, differences in sample types and ICU-specific data could explain the discrepancies compared to other studies.

Aztreonam, a β-lactam antibiotic with a single β-lactam ring is tier 4 drug as per CLSI [18, 19]. It is hydrolyzed by Class A (ESBLs, KPC) and Class C (AmpC) β-lactamases but remains stable against Class B (MBLs), allowing its use in combination with β-lactamase inhibitors like avibactam. In our study, aztreonam susceptibility remained low (50-60%) compared to ICMR data, except in 2021, when it peaked at 90%, aligning with the highest number of susceptible isolates that year [20]. Similar findings were reported by Bazghandi et al. (57%) and Krovvidi et al. (52%), while Reddy et al. documented a lower susceptibility rate of 25% [7, 29, 30].

The study constraints included its single-center, retrospective design, the exclusion of certain sample types (blood, CSF, and urine) and the unavailability of detailed patient demographic information.

The field of antimicrobial resistance is highly dynamic, thus posing continuous challenges for clinicians in managing resistant infections. Monitoring updated trends in the prevalence of MDR pathogens, such as *P. aeruginosa*, and their susceptibility profiles is essential for reviewing treatment strategies. This will aid in incorporating new evidence-based data to strengthen hospital surveillance and infection control practices within the institute and other settings with limited resources and high patient influx.

Conclusion

P. aeruginosa remains a significant gram-negative pathogen in hospital settings. Over the past four and a half years, its prevalence has significantly declined, with a more marked reduction following the implementation of stringent infection control measures. Improved susceptibility to piperacillin-tazobactam, carbapenems, aztreonam, and aminoglycosides was observed, although susceptibility to fluoroquinolones and monobactams remained low. As AMR continues to evolve, these updated dynamics in the susceptibility profiles of MDR pathogens are vital for optimizing treatment strategies and enhancing hospital surveillance and infection control measures.

Declarations

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Conflicts of interest: None to declare.

Ethics approval: As this study used retrospective laboratory data, individual patient consent was not required. Individual patient consent was not required, and formal Institutional Review Board approval or waiver was not applicable for this analysis.

Informed consent: Not applicable.

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