

Research Article

Efficacy of Antibiotics Against *Pseudomonas aeruginosa* Isolated from Respiratory Devices in Tanzania

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Abstract

Pseudomonas aeruginosa is an opportunistic pathogen responsible for severe hospital-acquired infections, including ventilator-associated pneumonia. Due to its biofilm-forming ability and antimicrobial resistance, it presents a significant public health challenge. This study assessed the efficacy of selected antibiotics against *P. aeruginosa* isolates associated with respiratory devices from regional referral hospitals in mainland Tanzania. A cross-sectional study was conducted in 2024, in which samples were collected from January to March in Emergency wards, Intensive care units, and Medical wards. Samples collected included water from the Oxygen humidification container, swab samples from reusable masks and the connectors. Laboratory analysis using standard microbiological techniques and PCR were employed for isolation and confirmation of *P. aeruginosa*. Antimicrobial susceptibility testing was performed using the Kirby-Bauer disc diffusion method. Out of the collected samples (N=231), *P. aeruginosa* was detected at an overall prevalence of 14.7% (n=34). The analysis of prevalence by sample type revealed that water for humidification had the highest prevalence of 30.6%, followed by respirators at 8.2% and the least in connectors at 3.5%. *P. aeruginosa* showed a notable resistance towards gentamycin, followed by meropenem, and the least resistance was shown in ceftazidime. On the other hand, *P. aeruginosa* were fully susceptible to piperacillin-tazobactam combination. A very small proportion of isolates demonstrated multidrug resistance (MDR). Despite the noted resistance majority of the antibiotics used to treat respiratory tract infection (RTIs) in this study showed significant efficacy.

Keywords

Pseudomonas aeruginosa Detection, Respiratory Tract Infection, Antibiotic Resistance, Oxygen Therapy Devices

1. Introduction

Pseudomonas aeruginosa is a prevalent opportunistic bacterium that leads to acute nosocomial necrotizing pneumonia

and is the primary cause of persistent lung infections in immunocompromised individuals [1]. They can grow well at

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25°C to 37°C under aerobic conditions and can be isolated from several environments such as water, soil, and mammal tissue [2]. Gram-negative bacteria like *P. aeruginosa*, amongst the *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Enterobacter spp.* (ESKAPE) group members are highly characterised for their antimicrobial resistance due to their ability to form a biofilm [3]. Effective healthcare organizations like the Center for Disease Control (CDC) and the World Health Organization (WHO) have identified this concern and recommended a response to the crisis [4]. A study in 2019 on antimicrobial resistance (AMR) revealed that out of the estimated 4.95 million deaths associated with AMR, 1.5 million deaths accounted for respiratory infections, which include among others contaminations in the respiratory devices by the *P. aeruginosa* which is a member of the ESKAPE group of pathogens [5]. Healthcare-associated infections pose significant challenges to patient safety and healthcare quality globally, with respiratory infections being particularly prevalent and severe. Studies across Europe, Asia, and America have reported that *P. aeruginosa* has been the main cause of several Hospital-acquired infections (HAIs), such as Ventilator Associated Pneumonia, while accounting for 20% of all HAIs in Europe and the United States [6].

Studies conducted in India reported that *P. aeruginosa* is the fourth most common cause of lower respiratory infections [6, 7]. Immunocompromised patients, such as HIV/AIDS patients, are the most affected individuals with bacterial pneumonia caused by *P. aeruginosa* which is more prevalent in hospital settings, this situation leads to an increased rate of mortality and morbidity in this group [8]. Factors like medical condi-

tions, such as HIV/AIDS, cancer, malnutrition, old age, genetic disorders, and medications like immunosuppressants, as used in organ transplants, act as causative agents to immunocompromised individuals [9]. In a study done between April and May 2023 at Muhimbili National Hospital in Tanzania, bacterial isolates from 856 patients with bloodstream infections (BSI) were obtained, and 43.28% of all isolates were ESKAPE pathogens which are predominant in hospitals and *P. aeruginosa* accounting for 4.77% [5]. It has been reported that *P. aeruginosa* contribute up to 8% of all HAIs, like pulmonary infections (tracheobronchitis), urinary tract infections (UTI), skin and soft tissue (burn wounds), bacteremia and endocarditis [10]. To ensure the safety and well-being of patients undergoing treatment in referral hospitals with the help of respiratory devices, it is critical to determine that the devices do not contribute to HAIs.

Tanzania Medicines and Medical Devices Authority (TMDA), is regulatory authority in Tanzania, mandated to ensure the quality, safety and effectiveness of medicines and medical devices used in clinical practices. Therefore, this study aimed to isolate *P. aeruginosa* in hospital settings and assess the efficacy of selected antibiotics against the isolates.

2. Methodology

2.1. Study Area

The study covered all 28 Regional Referral Hospitals (RRH) and (1) one zone hospital in mainland Tanzania. The location of each of the visited referral hospital is shown in Figure 1.

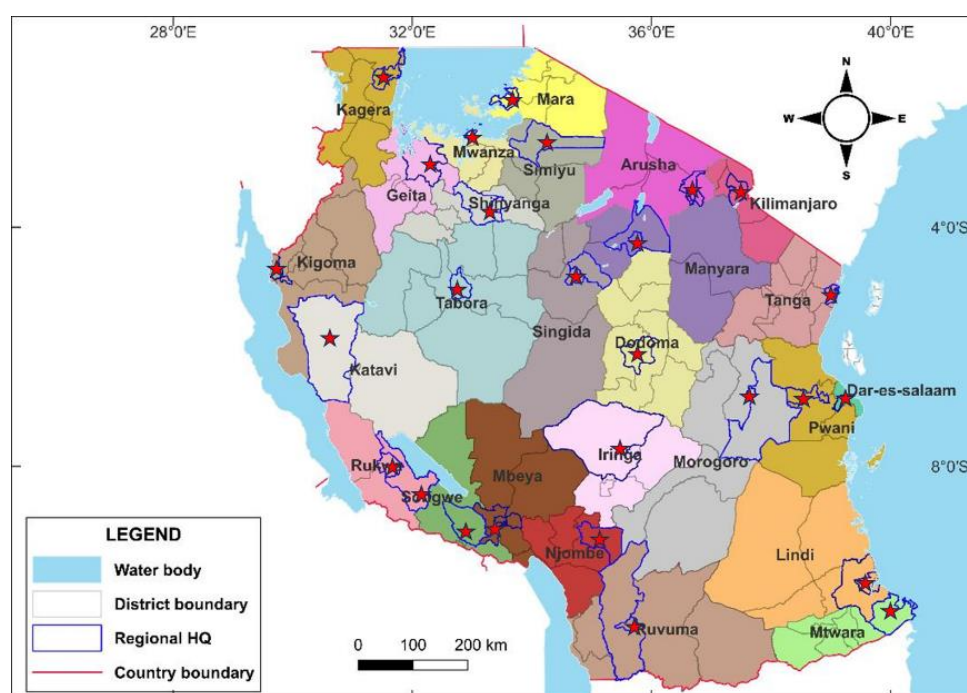


Figure 1. A map of Tanzania showing the locations of all RRH where samples were collected.

2.2. Study Design and Sampling Strategy

A cross-section study type and purposive sampling design (experiential criterion) was employed in this study between January and March 2024. The list of hospitals was obtained from the website of the Ministry of Health, Tanzania, <https://hfrs.moh.go.tz>. Names of all RRH visited were given a specific code for ease of tabulation and results presentation.

2.3. Sample Size and Determination

A total of 261 samples were expected from all RRH in Tanzania. These samples were to be collected from humidifiers, connectors, and reusable masks. Samples were gathered from three hospital departments namely; Emergency Units, Intensive Care Units, and Medical Wards.

2.4. Sample Collection

2.4.1. Water Samples from the Humidifier

Sterile Pasteur pipettes were used to aseptically collect water samples from a container used for oxygen humidification and then transferred aseptically to 15 mL sterile Falcon tubes. The falcon tubes were securely sealed and labelled, ready for transportation to the TMDA laboratory at a cool temperature of 2-8°C. A total of 85 water samples were obtained, consisting of 29 from the Emergency unit, 27 from the ICU and 29 from the medical wards.

2.4.2. Swab Samples

Swab samples from humidifiers, connectors and from masks or respirator or respirators were obtained by aseptically swabbing around the connector areas of the humidifiers and reusable oxygen masks. The swabs were then placed into transport media, sealed, labelled, maintained in cool temperature of 2-8°C and transported to TMDA laboratory for analyses within 24hrs.

2.5. Laboratory Analysis

2.5.1. Enrichment of Bacteria for the Isolation and Identification of *P. aeruginosa*

Collected samples were inoculated into nutrient broths to promote the multiplication of the microorganisms. Tryptic Soy Broth (TSB) (Oxoid UK) was used for water samples from humidifiers at a ratio of 1: 10 serial dilutions. The swab samples also inoculated separately into TSB. The inoculated broths were incubated at 37°C for 24 hrs. as previously described [11].

2.5.2. Macro Morphology Identification

The enriched inoculums were then sub-cultured onto Cetrimide agar (CA) (Oxoid-UK), a selective medium used for the isolation of *P. aeruginosa*. The inoculated media were then incubated in a calibrated incubator (Mettler Incubator, Germany) at 32-35°C for 18-24 hrs. After incubation time plates were observed for round, bluish-green with smooth surfaces to imply presumptive *P. aeruginosa* colonies. The vivid single colony were sub-cultured onto Nutrient agar (NA), (Oxoid UK) to obtain a pure culture for subsequent analytical tests.

2.5.3. Micro Morphology Identification

Overnight pure cultures of suspected isolates were subjected to Gram staining procedure whereby single pure colonies were smeared on the slides, heat-fixed, and stained by Gram stain as per the manufacturer's procedure. The stained slides were observed under microscope at x100 oil immersion for Gram-negative rod bacilli. Isolates were then subjected to a biochemical test for species identification.

2.5.4. Biochemical Identification

Presumptive *P. aeruginosa* isolates were then tested for presence of Cytochrome oxidase enzyme. Briefly, a sterile applicator sticks was used to pick single colony from an overnight pure culture onto the surface of the oxidase test strip and observed for deep blue coloration to indicate a positive oxidase test, which is a biochemical characteristic of *P. aeruginosa*.

2.5.5. Confirmation of *P. aeruginosa* by Molecular Methods

(i). Genomic DNA Extraction

The presumptive *P. aeruginosa* isolates that showed green color on Cetrimide agar and were oxidase-positive were then subjected to PCR confirmation. Boiling method was employed to extract genomic DNA from presumptive *P. aeruginosa* isolates. Briefly, four pure colonies were suspended in Eppendorf tube containing 100µL of nuclease-free water as described [11]. The tubes were heated at 95°C inside the water bath for 5 minutes and then transferred into the -20°C freezer for 10 minutes to lyse the cells. The tubes were then centrifuged at 10,000 rpm for one-minute, Sterile Dnase free tips and calibrated micropipette was used to draw 80µL of the supernatant, which contained genomic DNA (gDNA), and transferred it into a clean PCR tube.

(ii). PCR Analysis

Confirmation of *P. aeruginosa* was performed using a molecular technique for detecting the presence of the *P. aeruginosa Oprl gene* with 504 bp in the presumptive isolates. The forward (Oprl F-5' ATG GAA ATG CTG AAA TTC GGC-3'), and the reverse primers (3'-CTT CTT CAG CTC GAC GCG ACG-5') were used as previously described [12]. Master Mix

was prepared, each PCR tube contained 16µL of *nuclease*-free water, 0.5µL of forward primer, 0.5µL of reverse primer, and 5µL of premix. Finally, 3µL of DNA template was added, making a total of 25µL PCR reaction mixtures. The PCR amplification conditions shown in Table 1 were used for this study.

Table 1. PCR amplification conditions.

Condition	Temperature (°C)	Time (minutes)	No. of cycles
Initial denaturation	96	5	1
Denaturation	96	1	40
Annealing	55	1	40
Extension	72	1	40
Final extension	72	10	1

The PCR products were analyzed by gel electrophoresis whereby 1% agarose gel stained with ethidium bromide (0.5 µg/mL) was used. The presence and absence of DNA bands were visualized using a UV trans-illumination machine (UVP Bio-Doc-It imaging system, Cambridge-UK).

2.6. Antimicrobial Susceptibility Test

The antimicrobial susceptibility of the confirmed *P. aeruginosa* isolates was performed by using the Kirby-Bauer disc diffusion method. Mueller Hinton Agar (MHA) (Oxoid UK) lot no. 2114574 was used as described in the Clinical and Laboratory Standards Institute (CLSI) 34th Edition of performance standards for antimicrobial susceptibility testing [13]. Pure colonies were subjected to antimicrobial susceptibility test against antibiotics agent from five groups of a namely Cephalosporins, Fluoroquinolones, Aminoglycosides, Carbapenems, and β-lactam combination. Overnight pure culture of *P. aeruginosa* were suspended in Phosphate-Buffered Saline (PBS) to obtain a solution with a turbidity equivalent to a 0.5 McFarland standard solution corresponding to about 1.5×10^8 CFU/mL of bacterial concentration. Using a sterile swab, the suspended colonies were streaked onto Muller Hinton agar plates and allowed to dry. Antibiotic discs of Ceftazidime 30µg, Ciprofloxacin 5µg, Gentamicin 10µg Meropenem 10µg- Piperacillin/Tazobactam 100/10µg- (Liofichem, Italy) were placed on the inoculated agar surface, kept for 1 hour at room temperature to allow diffusion to take place and incubated at 37°C for 24 hrs. After incubation, the zones of inhibition were measured using calibrated vernier calliper and meas-

urements were recorded and interpreted as resistant, intermediate or susceptible according to CLSI Guidelines 34th Edition. MDR strains of *P. aeruginosa* were identified when isolates found to be resistant to more than three different classes of antibiotics. *P. aeruginosa* ATCC 27853 was used as a positive control for all stages requiring positive controls.

2.7. Quality Control

Sterility and verification procedures were employed as per [11] materials such as Falcon tubes and Pasteur pipettes were checked for sterility before use, similarly all culture media were also verified. Positive and negative controls were used for all samples received from the field to ensure the reliability of the results as previously described [14].

2.8. Statistical Analysis

Statistical analysis was conducted using Statistical Product and Service Solutions (SPSS) version 20.0. The Chi-square (χ^2) test was used to assess associations between categorical variables. A *p*-value <0.05 was considered statistically significant in the multivariate analysis.

3. Results

3.1. Sample Collection Responses

The collected samples amounted to 88.5% (n=231) of the expected samples (N=261) from all regional referral hospitals in Tanzania mainland. These samples included 85 samples from water for oxygen humidification, 85 from connectors and 61 from reusable masks. Samples were gathered from three hospital departments namely Emergency Units, Intensive Care Units and Medical Wards. The variation observed in sample numbers, specifically in reusable masks, was due to some hospitals use only disposable oxygen masks which allows only to be used once and then dispose of, few hospital facilities still use reusable masks after disinfecting them.

3.2. PCR Identification of *P. aeruginosa*

PCR amplification of the *OprI* gene target was successfully attained for most of the presumptive positive tested *P. aeruginosa* isolates, as demonstrated by the agarose gel electrophoresis image. Clear and distinct single bands of uniform molecular size (504bp) were observed across the majority of presumptive positive *P. aeruginosa* sample lanes. The absence of non-specific bands and smearing indicates high specificity of the primers and good DNA quality. Lanes that showed an absent band, suggests the absence of the *P. aeruginosa OprI* gene in the samples, as shown in Figure 2.

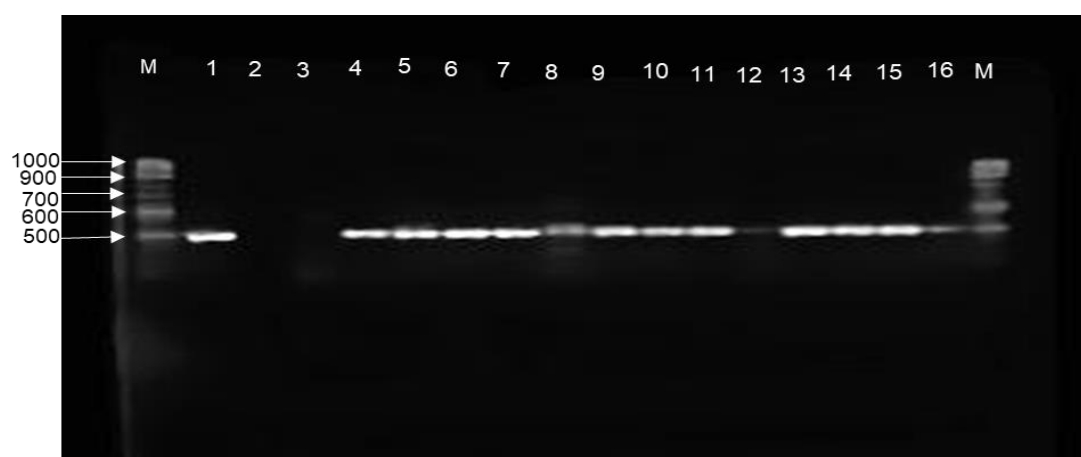


Figure 2. Agarose gel electrophoresis of PCR-amplified *Opnl* gene from *P. aeruginosa* isolates, lane M; markers, lane 1; positive control, lane 2; negative control and lane 3-16; isolate samples.

3.3. Prevalence of *P. aeruginosa* Across Hospitals

Among the 231 samples collected from regional referral hospitals, *P. aeruginosa* was detected in 34 of them, corresponding to an overall prevalence of 14.7%. The analysis of χ^2 and *p*-value revealed no significant difference in prevalence of *P. aeruginosa* across the hospitals ($\chi^2 = 25.637$, $p = 0.375$) as shown in Table 2. Similar trends in prevalence were also observed in other studies when the isolation was performed on clinical samples from critically ill patients [15, 16]. Looking

at individual facilities where samples were collected for this study, the highest prevalence per hospital was observed at 44% ($n=4$) out of the collected samples ($n=9$) per hospital facility. On the other hand, *P. aeruginosa* was not detected in samples from 9 hospitals, which is 31% of all hospitals, while 20 (69%) of the hospitals had at least one *P. aeruginosa*-positive sample. Results from other studies have shown the association of *P. aeruginosa* and respiratory devices used in hospital settings, which underlines the importance of these bacteria in treatment protocols [17]. These results suggest a varied approach and intensity of infections control and hospital disinfection approaches, and call for continued effort in the same direction to attain a unified HAIs control.

Table 2. Prevalence of *P. aeruginosa* across Hospitals.

Hospital codes	<i>P. aeruginosa</i>		χ^2	<i>p</i> -value
	Absent	Present		
	n (%)	n (%)		
RRH001	6 (85.7)	1 (14.3)	25.637	0.375
RRH002	8 (88.9)	1 (11.1)		
RRH003	7 (100)	0 (0)		
RRH004	5 (62.5)	3 (37.5)		
RRH005	6 (100)	0 (0)		
RRH006	6 (75)	2 (25)		
RRH007	9 (100)	0 (0)		
RRH008	6 (75)	2 (25)		
RRH009	8 (88.9)	1 (11.1)		
RRH010	5 (62.5)	3 (37.5)		
RRH011	4 (66.7)	2 (33.3)		

Hospital codes	<i>P. aeruginosa</i>		χ^2	<i>p</i> -value
	Absent	Present		
	n (%)	n (%)		
RRH012	7 (87.5)	1 (12.5)		
RRH013	9 (100)	0 (0)		
RRH014	9 (100)	0 (0)		
RRH015	6 (85.7)	1 (14.3)		
RRH016	9 (100)	0 (0)		
RRH017	6 (75)	2 (25)		
RRH018	7 (77.8)	2 (22.2)		
RRH019	8 (88.9)	1 (11.1)		
RRH020	8 (88.9)	1 (11.1)		
RRH021	7 (87.5)	1 (12.5)		
RRH022	7 (87.5)	1 (12.5)		
RRH023	7 (77.8)	2 (22.2)		
RRH024	6 (85.7)	1 (14.3)		
RRH025	5 (55.6)	4 (44.4)		
RRH026	5 (100.0)	0 (0)		
RRH027	7 (77.8)	2 (22.2)		
RRH028	6 (100)	0 (0)		
RRH029	8 (100)	0 (0)		
Total	197 (85.3)	34 (14.7)		

3.4. Distribution of *P. aeruginosa* Across the Hospital Units

The assessment of the isolation of *P. aeruginosa* revealed that, among the collected samples (N=231), 85.3% (n=197) were negative, while 14.7% (n=34) were positive for *P. aeruginosa*. This low positive rate specifically for *P. aeruginosa* signifies the effectiveness of hygiene and infection control in hospital settings as shown in Table 3. However, despite the low detection rate, the association of *P. aeruginosa* with respiratory devices in some facilities suggests inadequacy in infection control, which may be attributed to the types of disinfectants used and the frequency of disinfection. These results call for a detailed study on the efficacy of hospital disinfectants used. Studies elsewhere reported the association of *P. aeruginosa* having ability to form biofilm, thriving in harsh and wet environments, such as the oxygen humidification process which ultimately led to spreads of opportunistic infections and hence raises concerns [18].

Table 3. Distribution of *P. aeruginosa* across the hospital units.

Unit name	<i>P. aeruginosa</i> isolation [n (%)]			
	Positive	Negative	χ^2	<i>p</i> -value
EMD	11 (13.9)	68 (86.1)	1.371	0.523
ICU	14 (18.4)	62 (81.6)		
WARD	9 (11.8)	67 (88.2)		
TOTAL	34 (14.7)	197 (85.3)		

3.5. Distribution of the Positivity Rate of *P. aeruginosa* Across the Hospital Units

The distribution of *P. aeruginosa* across the hospital units showed that among the total positive isolates (N=34), 41.2%(n=14) were detected from the ICUs, while the EMD

ward accounted for 32.4% and 26.5% (n=9) in medical wards, respectively. Despite the slightly high prevalence in the ICU, generally, there was no significant difference in prevalence across the wards ($\chi^2 = 1.37$, $p = 0.5$) as shown in Table 4. The overall low prevalence could be explained by the effectiveness of decontamination implementation for infection control

measures in most sampled hospital environments. Additionally, isolation of *P. aeruginosa* in emergency wards suggests that HAIs with *P. aeruginosa* can begin as early as arrival in the EMD. These results depict that hospital hygiene and infections control need more stringent control since there is a very high interaction between the hospital environment and the incoming patients and their relatives.

Table 4. Distribution of positivity rate of *P. aeruginosa* across the hospital units.

Unit name	<i>P. aeruginosa</i> isolates		χ^2	P-value
	n	%		
EMD	11	32.3	1.371	0.523
ICU	14	41.2		
WARD	9	26.5		
TOTAL (N)	34	100		

3.6. Prevalence of *P. aeruginosa* per Sample Type

Prevalence of *P. aeruginosa* across sample types, namely connector (CS), water for humidification (WS) and reusable masks samples (RS), revealed that *P. aeruginosa* prevalence was notably high in WS 30.6% (n=26), followed by CS at 8.2% (n=5) and in RS 3.5% (n=3). However, the overall undetected rate was at 85.3% for all samples pooled together as shown in Table 5. These findings were predicted due to the ability of *P.*

aeruginosa to thrive in wet environments such as long-standing water or non-sterile water when used to humidify oxygen. This may present a high risk of introducing bacteria into already sick patients, resulting in complications to the treatment regimen. To make the matter worse, once bacteria are present in water for humidification, they may be distributed throughout the oxygen therapy system, including valves and the connector and possibly form biofilms which make them difficult to sanitize. Findings from another study conducted in Italy revealed a similar tendency, which shows bacterial contamination of the water from the humidifier from the medical ward, surgical, and emergency units were 83%, 77% and 50%, respectively [19].

Table 5. Prevalence of *P. aeruginosa* across sample types.

Sample type	<i>P. aeruginosa</i>		χ^2	p= value
	Absent n (%)	Present n (%)		
Connector (CS)	82 (96.5)	3 (3.5)	27.599	<0.0001
Water for humidification (WS)	59 (69.4)	26 (30.6)		
Respirator/reusable Masks (RS)	56 (91.8)	5 (8.2)		

3.7. Antimicrobial Susceptibility Test

The antimicrobial susceptibility of *P. aeruginosa* isolates

against six different antibiotics revealed that among the confirmed *P. aeruginosa* isolates (n=34) majority were susceptible to the six antibiotics commonly prescribed for treatment of RTIs in Tanzania. A small proportion were resistant to Gentamycin (CN) and Meropenem (MRP) and to a lesser extent

Ciprofloxacin (CIP) and Ceftazidime (CAZ). Piperacillin/Tazobactam (TZE) combination showed the highest efficacy against the clinical isolates used in this study as shown in Figure 3. Recent studies have highlighted the difficulties in treating critically ill patients. [16, 20] This study revealed *P. aeruginosa* being isolated in respiratory devices, in the ICU, where critically ill patients receive oxygen therapy, hence suggesting difficulties or treatment burden endured by patients. Furthermore, this bacterium has been reported to have a high mortality rate compared to other bacteria [21]. When linking

to the current findings in this study, it strongly calls for thorough hospital hygiene and sanitation, especially in low-income settings where access to medications is limited.

These results suggest the need to address the observed resistance, but generally highlight that the majority of currently available antibiotics recommended for the treatment of respiratory tract infection in Tanzania are still effective. Henceforth, we recommend ongoing rational use and AMR stewardship to protect these critically important medicines.

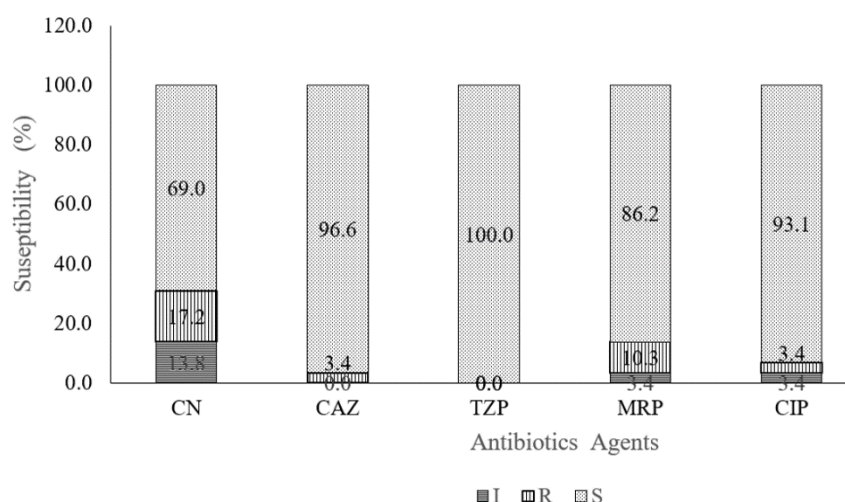


Figure 3. The Antimicrobial susceptibility profile of *P. aeruginosa* against six different antibiotics for treatment of RTIs in Tanzania.

3.8. Prevalence of Multidrug Resistance Among Isolates

Overall, the findings indicated a low prevalence of MDR among *P. aeruginosa* isolates across all regional referral hospitals, with only 11% (n = 4) from a single hospital identified as AMR, as shown in Table 6. This unique case presumes that

it could be the same isolate circulating in the same hospital. A bigger study with wide coverage to establish the extent of MDR, if any, at district hospitals. The difficulties in treating MDR have been reported in other studies [15], thus strict measures are required to limit their spread to other facilities. This finding further necessitates future studies to be conducted to isolate resistant genes and establish phylogenetic relationships among the isolates.

Table 6. Antibiotic Resistance Profiles and Status of *Pseudomonas* Isolates across Various Hospitals.

Hospital code	Susceptible (S) (n)	Intermediate (I) (n)	Resistance (R) (n)	MDR status
RRH001	5	0	1	Not MDR
RRH002	5	1	0	Not MDR
RRH003	2	0	4	MDR
RRH006	11	0	1	Not MDR
RRH008	5	1	0	Not MDR
RRH009	6	0	0	Not MDR
RRH010	24	0	0	Not MDR
RRH011	10	0	2	Not MDR

Hospital code	Susceptible (S) (n)	Intermediate (I) (n)	Resistance (R) (n)	MDR status
RRH012	6	0	0	Not MDR
RRH015	6	0	0	Not MDR
RRH018	12	0	0	Not MDR
RRH017	11	1	0	Not MDR
RRH020	6	0	0	Not MDR
RRH021	6	0	0	Not MDR
RRH022	4	0	2	Not MDR
RRH023	6	0	0	Not MDR
RRH024	5	1	0	Not MDR
RRH025	23	1	0	Not MDR
RRH027	5	1	0	Not MDR

4. Conclusion

The current study highlighted several critical findings; first most antibiotics used to treat RTIs are still efficacious in case of HAIs by the *P. aeruginosa*, second the antibiotic resistance was low in percentage, but worth noting. The presence of MDR isolates in one of the RRHs accentuates the challenges of managing infections with antimicrobial therapies and water for humidification showed significant bacterial colonization. The prevalence of these pathogens varied across different hospital units, with *P. aeruginosa* showing a higher presence in ICUs and EMDs and thus strict infection control measures are warranted.

Abbreviations

AMR	Antimicrobial Resistance
ATCC	American Type Culture Collection
CA	Cetrimide Agar
CDC	Center for Disease Control
CLSI	Clinical and Laboratory Standards Institute
EMD	Emergence Department
ESKAPE	<i>Enterococcus faecium</i> , <i>Staphylococcus aureus</i> , <i>Klebsiella pneumoniae</i> , <i>Acinetobacter baumannii</i> , <i>Pseudomonas aeruginosa</i> and <i>Enterobacter Spp.</i>
HAI	Hospital Acquired Infections
ICU	Intensive Care Unit
MDR	Multi Drug Resistance
MHA	Mueller Hinton Agar
PCR	Polymerase Chain Reaction
RRH	Regional Referral Hospital
NA	Nutrient Agar

RTI	Respiratory Tract Infection
SPP	Species
SPSS	Statistical Product and Service Solutions
TAE	Tris-acetate-EDTA
TMDA	Tanzania Medicines and Medical Devices Authority
TSB	Tryptose Soy Broth
USP	United State Pharmacopoeia
USPNF	United States Pharmacopoeia-National Formulary

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Author Contributions

Adelard Bartholomew Mtenga: Conceptualization, Data curation, Formal Analysis, Methodology, Supervision, Writing – original draft, Writing – review & editing

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Danstan Hipolite Shewiyo: Project administration, Supervision, Writing – review & editing

Data Availability Statement

The data supporting the outcome of this research work have been reported in this manuscript.

Conflicts of Interest

Authors declare no conflicts of interest.

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