American Journal of Infectious Diseases and Microbiology, 2023, Vol. 11, No. 1, 1-7
Available online at http://pubs.sciepub.com/ajidm/11/1/1
Published by Science and Education Publishing
DOI:10.12691/ajidm-11-1-1

Prevalence of Multidrug-Resistant, Extensively Drug-Resistant and Pandrug-Resistant *Pseudomonas aeruginosa* Clinical Isolates in Khartoum State, Sudan

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Received November 21, 2022; Revised December 28, 2022; Accepted January 09, 2023

Abstract **Background.** Pseudomonas aeruginosa is a common opportunistic Gram-negative pathogen responsible for a wide range of hospital acquired infections that may present high rates of antimicrobial resistance. It could also become multidrug-resistant (MDR), extensively drug-resistant (XDR), and pan drug-resistant (PDR) during a short period. The aim of the present study is to determine the prevalence of MDR, XDR and PDR-P. aeruginosa clinical isolates in Khartoum State-Sudan. **Materials and Methods.** A multihospital laboratory-based study was conducted to collect *P. aeruginosa* clinical isolates from various clinical specimen's culture during eighteen-month period from March 2020 to December 2021. The *P. aeruginosa* strains were reidentified by conventional biochemical methods and genotypically by amplification of 16S rRNA gene by Polymerase chain reaction (PCR) assay. Antimicrobial susceptibility testing was done by Kirby-Bauer disc diffusion method. MDR, XDR and PDR were determined according to new recommendation of the Clinical and Laboratory Standards Institute and the European Committee for Antimicrobial Susceptibility Testing. **Results.** Of 512 *P. aeruginosa* clinical isolates were recovered from various clinical specimen's culture, only 289(66.4%) of the isolates were confirmed as *P. aeruginosa* strains genotypically, out of those *P. aeruginosa* strains were categorized regarding antimicrobial resistance to 98(33.9%) MDR, 61(21.1%) XDR and 2(0.7%) PDR. The XDR strains were exhibited significant overall resistance to all used antibiotic classes, whereas MDR strains were insignificant resistant to fluoroquinolones and polymyxins. Patients prior of hospitalized for 1-2weeks, uses of companied antibiotics therapy with duration of one week and source of isolation from wound swab and blood specimen ($P < 0.05$), remained independently associated with an increased likelihood of antimicrobial resistant in clinical *P. aeruginosa* isolates. **Conclusion.** The study highlights the increase prevalence of MDR and XDR *P. aeruginosa* clinical isolates in both hospital and community settings, along with emerged two pandrug resistant strains. Thus, continuous antimicrobial resistance surveillance for this bacterium is necessary for guiding antimicrobial treatment and stewardship as well as based knowledge for future comparative studies.

Keywords: *pseudomonas aeruginosa, clinical isolates, multidrug-resistant, extensively drug-resistant, pan drug-resistant, Khartoum*

Cite This Article: Sara E Mohammed, Omnia M Hamid, Sababil S Ali, Mushal Allam, and A M Elhussein, "Prevalence of Multidrug-Resistant, Extensively Drug-Resistant and Pandrug-Resistant *Pseudomonas aeruginosa* Clinical Isolates in Khartoum State, Sudan." *American Journal of Infectious Diseases and Microbiology*, vol. 11, no. 1 (2023): 1-7. doi: 10.12691/ajidm-11-1-1.

1. Introduction

Pseudomonas aeruginosa is a non-fermentative gram-negative bacteria and is one of the serious opportunistic pathogens act in patients with weakened immune systems [1]. This organism commonly causes life-threatening community-acquired pneumonia, eye infection, hospital-acquired infections such as pneumonia, urinary tract infections, bacteremia severe burn wound infections, and other parts of the body after surgery; as well as chronic lung infections in patients with cystic fibrosis [1,2]. Commonly, *P. aeruginosa* is an extraordinary pathogen that is intrinsically resistant to a wide range of antimicrobial agents with the capacity to acquired resistance through chromosomal mutations or acquire extra-chromosomal materials from

surrounding environments to various classes of antibiotics [3,4].

Recently, the increasing dissemination rates of between 15% and 30% in some geographical areas of multidrug-resistant (MDR) and extensively drug-resistant (XDR) *P. aeruginosa* strains at community and hospital settings, become an urgent global issues of public health concern [5]. MDR exhibited antibiotic resistance to different antibiotic including antipseudomonal penicillins plus β-lactamase inhibitors, antipseudomonal cephalosporins, fluoroquinolones, tetracycline, and aminoglycosides [6]. Treatment with colistin or combination therapy has become the only remaining active antibiotics treatment and the last resort in terms of treatment for MDR-*P. aeruginosa* [7]. Therefore, MDR-*P. aeruginosa was* also suggested as being XDR, which refers to resistance to all antibiotics except to 1 or 2 antibiotic classes mainly colistin [7]. This situation is associated with worse outcomes such as increased morbidity and mortality, emergence reported of pandrug-resistant (PDR) strains which can attribute to limited effective antimicrobial options [6,8]. Despite there are published report on drugs-resistant *P. aeruginosa* strains covering the entire region [9-12] that provide early warnings of emerging threats. Though, there are still gaps or limited data on identify the prevalence of MDR, XDR, and PDR phenotypes, and keeping track of demographic data about clinical isolates for long–term resistance trends. The purpose of this study was to evaluate the prevalence of mentioned phenotypes and their antimicrobial resistance patterns in order to assist in selecting empiric antibiotic therapy and monitoring emerging drug resistance patterns.

2. Materials and Methods

2.1. Study Design and Setting

A multihospital laboratory-based, cross-sectional study was conducted between March 2020 to December 2021. Remnant *P. aeruginosa* isolates were recovered from various clinical specimen's culture (wound swab, blood, urine, sputum, ear swab, body fluid aspirate, urine catheter, Cerebrospinal fluid (CSF), soft tissue swab, pus aspirate, nasal swab, high vaginal swab (HVS), and oral swab) of either inpatients or outpatients who were diagnosed with various infections obtained from Microbiology Laboratory of three hospitals in Khartoum State. In this study, we re-identified all presumptive (manually characterized) *P. aeruginosa* isolates, and the demographic data of the isolates corresponding to patients' sex, age, type of specimen, patient's status and antibiotic used was anonymously recorded. The collected culture strains were inoculated on nutrient agar (Himedia Company, India) plates aseptically and labelled with serial No, then transported to the National University-Microbiology Laboratory and process immediately.

2.2. Identification of *P. aeruginosa* Isolates

In this study, we re-identified all presumptive (manually characterized) clinical isolates of *P. aeruginosa* by standard microbiological methods which are adopted from Bailey & Scott's Diagnostic Microbiology guideline based on general phenotypic methods (colonial morphology, oxidase positivity, motility, pigment production, grape-like odour, oxidative carbohydrate utilization, and growth at 42°C) [13]. To confirm the *P. aeruginosa* isolates to species level, the PCR assays were used based on 16S ribosomal DNA (rDNA) sequence primers (PA-SS-F-5'-GGGGGATCTTCGGACCTCA-3' and PA-SS-R-5'-TCCT TAGAGTGCCCACCCG-3') [9] product length of amplicon was 956 base pair. Genomic DNA was extracted from overnight cultures of *P. aeruginosa* by boiling. All identified *P. aeruginosa strains* were stored at −80°C in Tryptic Soy Broth (TSB) with 5% glycerol and genomic extracted DNA were stored in −20°C. These vials were labelled with our laboratory identification number.

2.3. Antimicrobial Susceptibility Testing

Disk diffusion method was performed for detection of antimicrobial susceptibility in clinical isolates of *P. aeruginosa*, using Muller-Hinton agar with 0.5 McFarland standard for adjusting inoculum to 10^5/CFU. The susceptibility was interpreted according to CLSI-2021 guidelines (CLSI, M100S 31th edition breakpoints) [14]. The antimicrobial discs including: gentamicin (10µg), tobramycin (10µg), amikacin (30µg); impenem (10µg), meropenem (10µg), ceftazidime (30 µg), cefepime (30µg), ciprofloxacin (5 µg), levofloxacin (5µg), ticarcillin-clavulanic acid (85 µg), piperacillin-tazobactam (110µg), aztreonam (30 µg), fosfomycin (200µg), and colistin (10µg). All these disks were obtained from HiMedia Laboratories (India). The diameter of inhibition zones was measured by (mm) according to (CLSI) and data report as resistance (R), Intermediate (I), or susceptible (S), as the susceptibility of the test organism is proportional to the zone of inhibition produced by the antibiotic used(14). Control strain used for all antibiotics disks was *P. aeruginosa* ATCC27853.

2.4. Detection of MDR, XDR and PDR Isolates

Defining of MDR, XDR and PDR in *P. aeruginosa* isolates were done according to new standardized international document [15] recommendation by CLSI and EUCAST. Therefore, isolates of *P. aeruginosa* , which have shown resistant to at least 1 agent in ≥ 3 antimicrobial categories considered MDR, isolates show resistant to at least one agent in ≥6 antimicrobial categories known as XDR, and isolates exhibits non-susceptible to all to all commercially available agents for empirical in treatment' in all antimicrobial categories known as PDR [15].

2.5. Data Analysis

Data were entered, coded and exported into Statistical Package for Social Science (SPSS) software program [version 24.0 computer program (SPSS, Inc., Chicago, IL, USA)]. The analysis of descriptive statistics was used to see the relationship between the dependent variable (antimicrobial resistance pattern of each *P. aeruginosa* isolate), and independent variables (age, sex, type of

specimen, patient's status, types of antimicrobial used for treatments, year of bacterial isolation and hospital region). Then the determined frequencies of different variables were compared and presented in words, tables, and figures. Where mean was calculated with standard deviation (SD) for continuous variables and Chi-square were performed. The p-value of ≤ 0.05 was set as the significance level of the study.

2.6. Ethical Approval

Permission for conducting this research was granted by the Ethical Committee of the Faculty of Graduate Studies and Scientific Research– National University-Sudan-PhD. degree in Medical Laboratory Sciences-Microbiology, Khartoum, Sudan 2020, and from general directors of hospitals. Ethical approval was obtained from the ethical review committee in State Ministry of Health and Sudan Federal Ministry of Health (FMOH) in Khartoum state, Sudan. Patient's sociodemographic data was extracted from hospital record upon permission of hospitals general directors.

3. Results

The study excluded 223 isolates belonged to either *Pseudomonas* spp, or other gram-negative species or which failed to grow on subculture. Of the remaining 289 (56.5%) isolates were identified as *P. aeruginosa*, to species level. The maximum numbers of isolates (n=209) were detected among (≥20-60 years) age groups with mean age of 41.8± 16.8 years and male study subjects were most commonly affected by *P. aeruginosa* infection with a rate of 61.9%. Isolates were recovered from 55.7% outpatients, followed by 27.0% inpatients (different wards) and 17.3% ICU patient's with 10.9±6.8 days' mean of hospitalization duration. They were collected from different clinical specimens, including wound swab 71 (24.6%), blood 62 (21.5%) urine 61(21.1%), sputum, ear swab, body fluid aspirate and urine catheter 33 (11.4%), 21(7.3%), 15(5.2%) and 11 (3.8%), respectively. And from 1.7% to 0.3% of them were from cerebrospinal fluid (CSF), soft tissue swab, pus aspirate, nasal swab, high vaginal swab (HVS) and oral swab.

The resistance profiles of 289 *P. aeruginosa* isolates against fourteen different types of tested antimicrobial agents was as follows: highly resistant to ticarcillin-clavulanic acid 89.2%, followed by meropenem 50.9%, cefepime and fosfomycin 37.7% for each, impenem 37.4%, ceftazidime 31.8%, aztreonam 25.3%, fluoroquinolones 23.5-21.4%, and gentamycin 20.8%. Whereas *P. aeruginosa* isolates were exhibited low resistant to tobramycin 14.5%, amikacin 11.4% and colistin 9.7% as represented in (Table 1). Out of 161 of *P. aeruginosa* isolates showed antimicrobial resistance profiles and categorized according to CLSI and EUCAST recommendation (15)as follow; 98 (33.9%) were MDR, 61(21.1%) were XDR, and 2 (0.7%) were PDR (Table 2).

Considering the relationship between antimicrobial resistance categories (MDR, XDR and PDR) with resistance to different antimicrobial classes; for MDR strains only fluoroquinolones and polymyxins B resistant strains were statistically insignificant ($P > 0.05$), for XDR strains all classes of antimicrobial agent's resistant strains were statistically significant ($P \leq 0.05$), whereas PDR strains aminoglycosides and monobactams resistant strains were statistically significant ($P \leq 0.05$) are presented in (Table 3).

Univariate analysis predicated variables (age, gender and clinical demographic characteristics) of patients were the clinical isolates were obtained; inpatient being hospitalized for 1-2weeks [OR = 2.53; 95% CI = 1.51-4.23; P=0.004; MDR=32; XDR=23], patient history of companied antibiotics therapy [OR=3.30; 95%CI=1.83-5.96; P<0.0001; MDR=23; XDR= 16], duration of antibiotics taken for 1 week [OR=1.83; 95%CI= 0.99-3.41; P=0.05; MDR=25; XDR=18], and colonization of *P. aeruginosa* in isolate sources wound swabs [OR=1.92; 95%CI=1.1-3.41; P= 0.025; MDR=25; XDR= 13; PDR=1] and blood [OR=1.94; 95%CI=1.08-3.51; P=0.027; MDR=23; XDR=12] remained independently associated with an increased likelihood of antimicrobial resistant in clinical *P. aeruginosa* isolates (Table 4). Whereas; the following factors were not associated with acquisition of drug resistant's (MDR, XDR, and PDR) *P. aeruginosa*: patient gender, age, hospital admission status, colonization of *P. aeruginosa* in isolate sources other than wound swab and blood preceding a diagnosis of bacteremia.

Table 1. Antimicrobial susceptibility patterns of *P. aeruginosa* (n=289) isolated from patients in Khartoum State; 2020-2021

Antimicrobial category	Antimicrobial agents	*P. aeruginosa* n (%)		
		S	I	R
Aminoglycosides	GEN (10µg)	222 (76.8%)	7 (2.4%)	60 (20.8%)
	TOB (10µg)	238 (82.4%)	9 (3.1%)	42 (14.5%)
	AK (30µ)	250 (86.5%)	6 (2.1%)	33 (11.4%)
Carbapenems	IMP (10µg)	164 (56.7%)	17 (5.9%)	108 (37.4%)
	MRP (10µg)	126 (43.6%)	16 (5.5%)	147 (50.9%)
Cephalosporins	CAZ (30µg)	149 (51.6%)	48 (16.6%)	92 (31.8%)
	CPM (30µg)	137 (47.4%)	44 (15.2%)	109 (37.7%)
Fluoroquinolones	CIP (5µg)	214 (74.1%)	13 (4.5%)	62 (21.4%)
	LE (5µg)	190 (65.7%)	31 (10.7%)	68 (23.5%)
Penicillins + β-lactamase inhibitors	TCC (75/10µg)	23 (8.0%)	8 (2.8%)	258 (89.2%)
	PIT (100/10µg)	125 (43.3%)	100 (34.6%)	64 (22.1%)
Monobactams	AT (30µg)	163 (56.4%)	53 (18.3%)	73 (25.3%)
Phosphonic acids	FO (200µg)	129 (44.6%)	51 (17.6%)	109 (37.7%)
Polymyxins	CL (10µg)	261 (90.3%)	-	28 (9.7%)

Notes: n: number of isolated *P. aeruginosa* strains; %: percentage of susceptibility patterns; S: sensitive; R: resistant; I: intermediate; Inhibition zone by mm according CLSI-2021; GEN: gentamicin; TOB: tobramycin; AK: amikacin; IPM: impenem; MER: Meropenem; CAZ: ceftazidime; CPM: cefepime; CIP: ciprofloxacin; LE: levofloxacin; TCC: ticarcillin-clavulanic acid; PIT: piperacillin-tazobactam; AT: aztreonam; FO: fosfomycin; CL: colistin.

Table 2. Distribution of MDR, XDR and PDR amongst P. aeruginosa strains studied (n=289)

Resistance profile	The most common resistance pattern			Number of antimicrobial resistance categories
Susceptible	B-lactams and β-lactamase inhibitor combinations: Ticarcillin-clavulanic acid and Piperacillin-tazobactam Cephalosporins: Ceftazidime and Cefepime			0-2
MDR	B-lactams and β-lactamase inhibitor combinations: Ticarcillin-clavulanic acid and Piperacillin-tazobactam Carbapenems: Imipenem and Meropenem Cephalosporins: Ceftazidime and Cefepime Fluoroquinolones: Ciprofloxacin and Levofloxacin Aminoglycosides: Gentamicin, Tobramycin and Amikacin			3-4
XDR	B-lactams and β-lactamase inhibitor combinations: Ticarcillin-clavulanic acid and Piperacillin-tazobactam Carbapenems: Imipenem and Meropenem Cephalosporins: Ceftazidime and Cefepime Fluoroquinolones: Ciprofloxacin and Levofloxacin Monobactams: Aztreonam Phosphonic acids: Fosfomycin Polymyxins: Colistin Aminoglycosides: Gentamicin, Tobramycin and Amikacin			≥6
PDR	B-lactams and β-lactamase inhibitor combinations: Ticarcillin-clavulanic acid and Piperacillin-tazobactam Carbapenems: Imipenem and Meropenem Cephalosporins: Ceftazidime and Cefepime Fluoroquinolones: Ciprofloxacin and Levofloxacin Monobactams: Aztreonam Phosphonic acids: Fosfomycin Polymyxins: Colistin			≥12 listed antibiotics
Total no.(%)	Susceptible	128	(44.3%)	
	MDR	98	(33.9%)	
	XDR	61	(21.1%)	
	PDR	2	(0.7%)	

Notes: MDR: Multidrug-Resistant; EDR: Extensive Drug-Resistant; PDR: Pan Drug-Resistant.

Table 3. Distribution of *P. aeruginosa* MDR, XDR and PDR (n=161) strains with total antimicrobial classes resistance

Total antimicrobial resistance classes	Antimicrobial resistance categories		
	MDR (n=98) n(%)	**XDR (n=61) n(%)**	**PDR (n=2) n(%)**
Aminoglycosides (n=62)	11 (17.7)	46 (74.2)	2 (3.2)
***P* value**	0.002	<0.0001	0.043
Carbapenems (n=159)	72 (45.3)	56 (35.2)	2 (1.3)
***P* value**	<0.0001	<0.0001	0.199*
Cephalosporins (n=128)	53 (41.4)	60 (46.9)	2 (1.6)
***P* value**	0.016	<0.0001	0.111*
Fluoroquinolones (n=72)	26 (36.1)	42 (58.3)	2 (2.8)
***P* value**	0.649*	<0.0001	.061*
Penicillins + β-lactamase inhibitors (n=258)	94 (36.4)	60 (23.3)	2 (0.8)
***P* value**	0.012	0.012	0.579*
Monobactams (n=73)	17 (23.3)	46 (63.0)	2 (2.7)
***P* value**	0.027	<0.0001	0.035
Phosphonic acids (n=109)	49 (45.0)	31 (28.4)	2 (1.8)
***P* value**	0.002	0.019	0.069*
Polymyxins B (n=28)	7 (25.0)	16 (57.1)	2 (7.1)
***P* value**	0.344*	<0.0001	0.050

Notes: MDR: Multidrug-Resistant; XDR: Extensive Drug-Resistant; PDR: Pan Drug-Resistant.

Table 4. Frequency of MDR, XDR, PDR and non-resistant *P. aeruginosa* strains regarding characteristics of patients:

Patient characters	R-PA (n=161)			S-PA (n=128)	*P*-value[1]	OR[2] (95% CI[3])
	MDR[(n=98)]	XDR [(n=61)]	PDR [(n=2)]			
Gender						
Male (n=179)	59	39	1	80	**0.932**	**0.98(0.68-1.43)**
Female (n=110)	39	22	1	48	**0.906**	**1.03(0.66-1.60)**
Age group						
<20 years (n=37)	12	4	0	21	**0.785**	**0.61(0.30-1.21)**
20-60 years (n=209)	72	46	1	90	**0.155**	**1.05(0.73-1.51)**
≥61 years (n=43)	14	11	1	17	**0.558**	**1.22(0.63-2.34)**

Patient characters	R-PA (n=161)			S-PA (n=128)	*P*-value[1]	OR[2] (95% CI[3])
	MDR(n=98)	XDR (n=61)	PDR (n=2)			
Status						
Inpatients (n=78)	32	14	0	32	**0.606**	**1.14(0.69-2.01)**
ICU patients (n=50)	12	15	1	22	**0.969**	**1.09(0.58-2.07)**
Outpatients (n=161)	54	32	1	74	**0.733**	**0.94(0.64-1.38)**
Hospitalization duration/weeks						
Non-hospital admission (n=163)	54	33	1	75	**0.724**	**0.93(0.63-1.37)**
Less than week (n=23)	7	1	1	14	**0.631**	**1.24(0.52-2.95)**
1-2 weeks (n=87)	32	23	0	32	**0.004***	**2.53(1.51-4.23)**
>2 weeks (n=16)	5	4	0	7	**0.353**	**0.61(0.22-1.71)**
Antibiotics taken 90 days prior to						
Untaken (n=163)	54	33	1	75	**0.724**	**0.93(0.63-1.37)**
Monotherapy (n=63)	21	12	1	29	**0.677**	**0.92(0.63-1.35)**
Companied therapy (n=63)	23	16	0	24	**<0.0001***	**3.30(1.83-5.96)**
Duration of taken antibiotics/weeks						
Less than week (n=61)	19	10	2	30	**0.922**	**0.97(0.53-1.79)**
1 week (n=65)	25	18	0	22	**0.050***	**1.83(0.99-3.41)**
Source of the clinical isolates						
Wound Swab (n=71)	25	13	1	32	**0.025***	**1.92(1.10-3.41)**
Blood (n=62)	23	12	0	27	**0.027***	**1.94(1.08-3.51)**
Urine (n=61)	17	16	0	28	**0.061**	**1.76(0.97-3.18)**
Sputum (n=33)	9	10	0	14	**0.109**	**1.83(0.87-3.83)**
Ear swab (n=21)	11	3	0	7	**0.377**	**1.52(0.59-3.92)**
Body Fluid aspirate (n=15)	7	1	0	7	**0.818**	**0.89(0.31-2.51)**
Urine Catheter (n=11)	2	2	1	6	**0.485**	**0.65(0.19-2.18)**
Cerebrospinal fluid (n=5)	3	2	0	0	**0.145**	**3.94(0.46-34.18)**
Tissue (n=4)	1	0	0	3	**0.457**	**0.42(0.04-4.11)**
Pus (n=2)	0	1	0	1	**0.870**	**1.26(0.08-20.34)**
Nasal swab (n=2)	0	1	0	1	**0.870**	**1.26(0.08-20.34)**
High vaginal swab (n=1)	0	0	0	1	**0.415**	**0.26(0.01-6.52)**
Oral swab (n=1)	0	0	0	1	**0.415**	**0.26(0.01-6.52)**

Notes: R-PA: antimicrobial resistant *P. aeruginosa;* MDR: Multidrug-Resistant; XDR: Extensive Drug-Resistant; PDR: Pan Drug-Resistant; S-PA: antimicrobial susceptible *P. aeruginosa*; 1. P* value <0.05 => statistical significance; 2. Odd Ratio, proportion of contribution to explain the outcome variable; 3. 95% confidence interval for the odd ratio.

4. Discussion

AMR is today a serious worldwide threat to the public health problems of the 21st century, due to the new resistance mechanisms spreading globally. Of further concern, it is now estimated that 700,000 people die each year as the result of drug-resistant infections, with this number predicted to increase to over 10 million deaths per year by 2050 [16]. In recent decades, *P. aeruginosa* is one of three critical pathogens contributing to the burden of antimicrobial resistant (AMR) in 2017 have been identified as priority pathogens by the World Health Organization (WHO) [17] and infections caused by *P. aeruginosa* are frequently life threatening and often difficult to treat due to the intrinsic resistance of *P. aeruginosa* to many antimicrobial agents. Furthermore, the resistance to antipseudomonal antibiotics has become an increasing problem in recent years [8,17], with the emergence of antimicrobial resistance during therapy occurring with a relatively high frequency [4,7].

In the present study, five hundred and twelve Gram-negative clinical isolates identified as *P. aeruginosa* by the different hospitals laboratory Khartoum State, 289 (56.5%) were confirmed according to genotypic methods (16S rDNA) to spices level. Genetic-based identification of *P. aeruginosa* from other Pseudomonas species is recommended by [18,19] as fundamental to efforts an accurate epidemiological picture of *P. aeruginosa* infection, to develop and assess new therapeutic strategies, whereas misidentification may contribute to wrong administration of antibiotics to patients.

In this study, a prevalence of 56.5% of *P. aeruginosa* strains found among [outpatients n=161; inpatients hospitalized in different wards n=78; ICU patients n=50] clinical isolates across Khartoum State hospitals laboratory. This frequency was higher to that reported in previous studies, in Khartoum which were (48.1%) in 2013 [20] and (24.7%) in 2018 [3]. Moreover; our study showed that 24.6% of *P. aeruginosa* isolated from wound swab the most common site of infection, followed by 21.5% from blood, 21.1% from urine, and 11.4%, 7.3%, 5.2% and 11 3.8% from sputum, ear swab, body fluid aspirate and urine catheter, respectively. And from 1.7% to 0.3% of them were from cerebrospinal fluid (CSF), soft tissue swab, pus aspirate, nasal swab, high vaginal swab (HVS) and oral swab. This was not in line with previous studies [12,21] in terms of sites order, but in general these sites are the most sites infected by *P. aeruginosa.* This different variation in *P. aeruginosa* prevalence which may be explained by the type and size of clinical specimens, the population studied, and hospital conditions.

Of 289 isolates of *P. aeruginosa* were tested for antimicrobial susceptibility against 14 agents from 8 antimicrobial categories. The current study confirms high prevalence of antibiotic of antibiotic-resistant *P. aeruginosa* in various clinical samples toward ticarcillin-clavulanic acid (89.3%), meropenem (50.9%) and fosfomycin (37.7%). Resistance to antipseudomonal

antibiotics range from 37.7% to 14.5% of *P. aeruginosa* strains were resistant to cefepime (37.7%), impenem (37.4%), aztreonam (25.3%), levofloxacin (23.5%), piperacillin-tazobactam (22,1%) and tobramycin (14.5%). There has been resistance to antipseudomonal drugs among strains worldwide and this poses a serious therapeutic problem when treating disease caused by these organisms [10], regardless in our finding it remain with good effectiveness. Overall, the highest susceptibility was shown for polymyxins and aminoglycosides antimicrobials (90.3%, 86.5% and 82.4% respectively, for colistin, amikacin and tobramycin). This finding in our study is consistent with studies from Sudan [11,12,22], Egypt [23], South Africa [24] and was more than study from Iran [21].

The current study, 98 isolates (33.9%) was categorized as MDR, 61 isolates (21.1%) as XDR and isolates (0.7%) as PDR among 289 clinical isolates of P. aeruginosa isolated from patients (inpatients and outpatients) across Khartoum State hospitals laboratory. The prevalence of MDR- *P. aeruginosa* varies widely across the globe, and our findings in this study are consistent with those observed in early 2018 Sudan [3], south Africa [25], Greece, Italy and Spain [26], and Lower than studies in Lebanon [27], Jordan [28], and Iran [29]. Furthermore, our finding is higher than in Qatar [30] and in China [31]. The high CPE prevalence in Sudan may be due to a variety of causes, including, unrestricted use of over-the-counter antibiotics or high selective pressures on the use of β-lactam antibiotics as first-line treatment for bacterial infections. Regarding the prevalence of XDR-*P. aeruginosa* our findings in this study are consistent with study done in Thailand [32], in Iran [29] and from Greece, Italy and Spain [26]. in addition; the alarming of two PDR- *P. aeruginosa* isolates, that were also resistant to colistin, our result is comparable other studies reported, the frequency of 3.8% (n=2) in Greece Italy and Spain [26], and 2.4% (n=5) in Qatar [30]. Contrary to what other studies reported, there was no PDR-*P. aeruginosa* isolates reported by Iran [29]. Consequently, one of the strengths of this study is the categorization of antimicrobial resistant profiles of *P. aeruginosa* registry, which may help develop an integrated database program that tracks every isolate of drug resistance in the future.

Our finding showed a significant correlation between MDR-*P. aeruginosa* strains and all used antimicrobial classes except fluoroquinolones class, and polymyxins class (P>0.05), furthermore the XDR-*P. aeruginosa* strains resistance profiles significantly related to all tested antimicrobials classes (P<0.05). While, an insignificant correlation between PDR-*P. aeruginosa* strains toward all used antimicrobial classes (P>0.05) except for aminoglycosides, monobactams and Polymyxins, (P=0.043, 0.035 and 0.050; respectively). These results of our study are similar with the findings of [15,21,30]. Understanding the antimicrobial resistance profiles of MDR, XDR or XDR-*P. aeruginosa* might be necessary to overcome infections with these resistant pathogens and to implement effective empirical antimicrobial treatments.

By univariate analysis, our study identified several risk factors significantly (P value <0.05) associated with antimicrobial resistance (MDR, XDR and PDR)-*P. aeruginosa* infection such as; inpatients hospitalized from 1-2 weeks, antibiotics companied therapy, duration of 1 week antibiotics using, wound Swab and blood source of isolates which increase risk from 1.21 to 5.96 times among patients in our sitting. The recent reporting of the introduction and spread of MDR and XDR-*P. aeurogenosa* associated infection into healthcare environments has also highlighted the invasive interventions, such as urinary catheters, drainage devices, mechanical ventilation, or renal dialysis, did pose a significant risk factor of patient [11,21,30,33]. Several surveys from developed and developing countries confirmed the direct relation between the irrational antibiotics use and the spread of resistant strains. To reduce this problem, it is very useful to implement proper strategy for infection control for example, good hygiene for hand and judicious use of antimicrobial antibiotic [9,21,34,35,36].

5. Conclusion

The prevalence of (33.9%) MDR, (21.1%) XDR, and alarming two PDR in clinical isolates of *P. aeruginosa* may highlight potential healthcare problems in the future, since we need to establish a standard protocol to identify clinical pathogens accurately to avoid false perceptions of their prevalence, along with continuous antimicrobial susceptibility monitoring**.**

Availability of Data and Materials

Upon request, the corresponding author can provide the datasets used during this study.

Competing Interests

No conflict of interest is declared by the authors**.**

Funding

No fund.

Authors' Contribution

In this study, (XXX) conceptualized the research idea and collected data; (XXX) analysed and interpreted the data; and (XXX) drafted the manuscript. All authors revised and approved the final manuscript.

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