

<https://doi.org/10.48047/AFJBS.6.12.2024.799-812>



African Journal of Biological Sciences

Journal homepage: <http://www.afjbs.com>



Research Paper

Open Access

Genotypic and Phenotypic Characterization of *Acinetobacter baumannii* Isolated From critically ill Patients in two healthcare facilities in Ebonyi State, Nigeria

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Article History

Volume 6 Issue 12, 2024

Received: 25 May 2024

Accepted : 25 June 2024

doi:

[10.48047/AFJBS.6.12.2024.799-812](https://doi.org/10.48047/AFJBS.6.12.2024.799-812)

ABSTRACT

Acinetobacter baumannii is one of the hospital acquired infections causing serious public health challenge in Nigeria. The present study was to determine the prevalence and antibiogram pattern of *Acinetobacter baumannii* from two public healthcare facilities Ebonyi State. A total of three hundred clinical samples were collected analyzed using standard microbiological procedures. All suspected bacterial isolates were phenotypically screened and positive results were subjected to antibiotic resistance test using the Kirby–Bauer disc diffusion technique and Multiple Antibiotic Resistance Index (MARI). Suspected *A. baumannii* isolates were finally confirmed using 16S rRNA sequencing. Of all the three hundred clinical samples studied, 20(9.0 %) samples from AE-FUTHA and 1 (1.25 %) sample from Mater Misericordia Hospital were

infected with *A. baumannii*. Catheter urine (16 %) from AE-FUTHA and (2 %) from Mater was the commonest sample sites for isolation of *A. baumannii* pathogen followed by wound sores (9 %) from AE-FUTHA. *A. baumannii* strains showed the highest level of resistance against tetracycline (100 %), trimethoprim

sulphamethoxazole, (100 %), ceftriaxone (80 %) and amikacin (80 %), and lowest resistance was against meropenem (14.2 %), imipenem (19.0 %) followed by polymyxin B (33.3 %), antibiotics level. The result of Multiple Antibiotic Index (MARI) showed a total 12.1 and an average of 0.57. Early diagnosis of infection caused by *A. baumannii* and its treatment with meropenem, imipenem or polymyxin B can reduce the mortality and morbidity risks of *A. baumannii* infection in critically ill patients.

Key: *Acinetobacter baumannii*, Antibiogram, Antibiotic resistance, Ebonyi State

INTRODUCTION

Acinetobacter baumannii is a very important pathogen that poses a substantial threat to patients. Its ability to resist routinely used antibiotics and its potential to cause severe infections in hospitals make it a major concern for healthcare professionals (Chao *et al.*, 2018). *Acinetobacter* is a Gram-negative bacterium that falls under the broader category of Gammaproteobacteria (Bitrian *et al.*, 2013). This assemblage of organisms consists of aerobic Gram-negative bacilli that have the ability to last for extended durations in the surroundings and on the hands of healthcare personnel (Zelege *et al.*, 2021). *Acinetobacter* is a medically significant bacteria that has extensive resistance to multiple medications. *Acinetobacter baumannii* is a significant cause of infection in vulnerable patients in the hospital (Bitrian *et al.*, 2013). *A. baumannii* is recognised as a highly opportunistic pathogen that significantly impacts individuals who are already weakened, particularly those in intensive care units (ICUs) and those with pre-existing illnesses. This organism regularly undermines the effectiveness of numerous antibiotics (Egwu *et al.*, 2021).

Despite initially being regarded as a pathogen with moderate virulence, subsequent research has demonstrated that *A. baumannii* is now recognised as one of the most important clinical pathogens (Chao *et al.*, 2018; Natalia *et al.*, 2018; Egwu *et al.*, 2021). Pneumonia has emerged as the primary symptom of nosocomial infections caused by this virus, leading to a substantial impact on the mortality rate of patients. This microorganism has also been identified as the causative agent responsible for a variety of other infections, such as septicemia, meningitis, urinary tract infection, endocarditis, septic shock, acute respiratory sore throat, and more recently, severe and fatal cases of necrotizing fasciitis (Gaddy *et al.*, 2012). In addition, *A. baumannii* is classified as one of the six ESKAPE pathogens (*Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Enterobacter species*) by the Infectious Diseases Society of America (IDSA). Shortly after, it rapidly evolved into a pan-drug resistant pathogen and has subsequently gained swift attention as one of the most significant bacterial pathogens for healthcare-associated infections (HAIs) (Falagas *et al.*, 2007). *A. baumannii* that is resistant to antibiotics has become a significant and challenging infection acquired in hospitals globally. The natural resilience of *A. baumannii* to thrive in harsh environmental circumstances and its capacity to withstand routinely employed medications in healthcare settings have solidified its position as a leading source of hospital-acquired infections (Muhammed, 2016). Due to the lack of thorough data in the current study region, the study aimed to specifically isolate and identify *Acinetobacter baumannii* from critically ill patients in two major healthcare facilities in Ebonyi State.

MATERIALS AND METHODS

Study area

The research was carried out at Alex Ekwueme-Federal University Teaching Hospital, Abakaliki (AE-FUTHA), and Mater Misericordia Hospital Afikpo, Ebonyi State, Nigeria. Ebonyi State is situated in the southeastern region of Nigeria, between longitude 7.30' and 8.30'E and latitude 5.40' and 6.45'N. The State was established on October 1, 1996, through the amalgamation of the old Abia and Enugu States, with Abakaliki serving as its capital. It is bordered to the north by Benue State, to the west by Enugu State, to the east by Cross River State, and to the south by Abia State. The State consists of thirteen Local Government Areas (LGAs), specifically Abakaliki, Ebonyi, Ishielu, Ohaukwu, Izzi, Ikwo, Ezza North, Ezza South, Afikpo North, Afikpo South, Ivo, Ohaozara, and Onicha LGAs. Several government and a few private health clinics are available in the study region.

Study design and period

This study was hospital-based cross-sectional design involving 300 critically ill patients on admission in medical, surgical, and orthopedic wards, and intensive care unit (ICU) of AE-FUTHA and Mater Misericordiae Hospital Afikpo. The study was conducted over a period of 6 months (1stSeptember, 2022 – 1st March, 2023)

Ethical clearance

Approval was granted by the Research and Ethical Committee (REC) of Alex Ekwueme-Federal University Teaching Hospital, Abakaliki (AE-FUTHA) under reference number AE-FUTHA/REC/VOL3/2022/129. Additionally, formal consent was received from the authorities of Mater Misericordiae Hospital Afikpo, Ebonyi State. Participation in this study was entirely optional, and participants who gave their agreement were included in the study. They had the freedom to withdraw from participation at any time without needing to provide reasons. Patients, as well as the spouse, parents, or carer of each participant, were approached and given verbal agreement. Subsequently, pertinent socio-demographic data and other essential information about the patient were acquired from the carer. The patient's demographic information, including age, gender, marital status, length of hospital stay, and the ward they were admitted to, was obtained and documented systematically.

Sample size and participant selection:

The sample size was determined using the Kish (1965), which yielded a minimum required number of participants of 300. The study included very ill patients who had been admitted to various hospital wards, including medical, surgical, orthopaedic, burns, and intensive care units (ICU), for a minimum of 14 days. The participants were selected using a sequential sampling approach throughout the study duration.

Data and sample collection

Different clinical samples were collected using sterile urine container and sterile swab sticks. The clinical samples collected include catheter urine (70), wound sores (67), skin swab (13), nasal swabs (20), wound drain (24) and oral swabs (26). All samples were collected aseptically and transferred to the Applied Microbiology Laboratory Unit of Ebonyi State University (EBSU), for further analysis.

Characterization and Identification of *Acinetobacter* Isolates

Bacterial colonies were identified using conventional microbiological culture-based assays, such as Gram-staining, catalase testing (with 3% hydrogen peroxide), indole, oxidase, coagulase, citrate, urease, Voges-Proskauer, DNase test, sugar fermentation, and the oxidation and fermentation of mannitol salt agar (16). The tests were conducted in accordance with established protocols. *Acinetobacter baumannii* ATCC1605 served as the positive control for every test protocol.

Microbial susceptibility testing

The antibiotic susceptibility of the isolates was evaluated using the disc diffusion method, as previously described by Onuoha *et al.* (2016). The suspension of colonies from each overnight culture was made using nutrient broth and then compared to the turbidity of 0.5 McFarland standards. The test organism was injected into Mueller-Hinton agar plates using a sterile swab stick. A 0.5 McFarland standard was used for the inoculation. The plates were then left undisturbed for 30 minutes for pre-diffusion. Discs containing antibiotics such as ampicillin, tetracycline, ciprofloxacin, ceftazidime, cefepime, gentamycin, meropenem, cefoperazone-sulbactam, doxycycline, imipenem, meropenem, levofloxacin, oxacillin, polymyxin B, and amikacin (Oxoid, UK) were placed on the media's surface and incubated at a temperature of 37 degrees Celsius for a duration of 24 hours. The diameter of the inhibition zones was measured using a metre rule, and the isolates were categorised as either susceptible or resistant based on the guidelines provided by the Clinical and Laboratory Standards Institute (CLSI, 2019). Multi-drug resistance (MDR) *A. baumannii* refers to *A. baumannii* that is resistant to three or more classes of antibiotics, including extended-spectrum cephalosporins (ceftazidime and cefepime), quinolones (ciprofloxacin and ofloxacin), aminoglycosides (amikacin and gentamicin), the combination of β -lactamase and β -lactamase inhibitors (ampicillin-sulbactam), and the carbapenems (ertapenem, imipenem, and meropenem).

Determination of Multiple Antibiotic Resistance (MAR) Index

Multiple antibiotic resistance (MAR) index was calculated and interpreted according to the method described by Ayandele *et al.*,(2020) using the formula; $MARI = a/b$, where 'a' is the number of antibiotics to which a particular isolate is resistant to and, 'b' is the total number of antibiotics tested.

DNA Extraction

The genomic DNA of the isolates was obtained by extracting it using a High Pure PCR Template Preparation Kit from Roche, located in Mannheim, Germany. Individual bacterial samples were transferred into properly labelled Eppendorf tubes for the purpose of extracting DNA. Adhering to the guidelines provided by the manufacturer. The PCR amplification process was conducted to confirm the presence of *A. baumannii* using the thermal cycler GeneAmp 9700 PCR System Thermalcycler (Applied Biosystem Inc., USA). The amplification targeted the 16S rRNA gene, using the forward primer 27f (5'-AGAGTTTGATCMTGGCTCAG-3') and the reverse primer 1525r (5'-AAGGAGGTGWTCCARCC-3'). The reaction mixture consisted of 0.3 units of Taq DNA polymerase (Promega, USA), which was diluted to a final volume of 12.5 μ l with sterile distilled water. Additionally, 6.44 μ l of DNA template was included. The PCR was conducted using a GeneAmp 9700 PCR System Thermalcycler (Applied Biosystem Inc., USA). The PCR profile included an initial denaturation step at 94°C for 5 minutes, followed by 30 cycles consisting of denaturation at 94°C for 30 seconds, primer annealing at 56°C for 30 seconds, and extension at

72°C for 1 minute and 30 seconds. The PCR was terminated with a final extension step at 72°C for 10 minutes. And relax at a temperature of 4 degrees Celsius.

Agarose gel electrophoresis

The DNA and PCR amplification were assessed for integrity using a 1.5% agarose gel. A solution was produced by combining 1.5% agarose in TAE (Trisacetate-EDTA-buffer) with 3 µl of ethidium bromide at a concentration of 0.5 g/ml. Subsequently, a DNA ladder (100 base pairs) was individually added to each well of the gel. Subsequently, the gel underwent electrophoresis for a minimum duration of one hour. The gel underwent electrophoresis at a voltage of 120V for a duration of 45 minutes. It was then observed using UV trans-illumination and captured in a photograph. The presence of the PCR product with the anticipated size of 789bp was verified using the Agarose gel electrophoresis technique

PCR product sequencing

The DNA products were sequenced using an Applied Biosystems Genetic Analyzer 3500 sequencer, following the instructions provided by the manufacturer. The sequencing kit used was the BigDye terminator v3.1 cycle sequencing kit. The sequences were compared to nucleotide sequences in GenBank using the BLAST programme from NCBI to determine the organism by identifying the most comparable 16S rRNA gene.

RESULTS

A total of 300 critically ill patients (220 from AE-FUTHA and 80 from MMHA) were recruited and clinical samples collected from each of them from the different wards of the two hospitals. A total of 21 (7.0%) participant samples were positive for *A. baumannii* with 20 (9.0%) of the 220 participants from AE-FUTHA) and 1(1.25%) of the 80 participants from MMHA being positive (Table 1).

Table 1: Frequency Distribution of critically ill patients with *Acinetobacter baumannii* infections at AE-FUTHA and Mater Misericordiae Hospital

Sample site	No of participants	No positive for <i>A. baumannii</i> (%)	No Negative for <i>A. baumannii</i> (%)
AE-FUTHA	220	20(9.0)	200(90.9)
Matter Hospital	80	1(1.25)	79 (98.7)
Total	300	21 (7.0)	279 (93.0)

Table 2 showed that the age of the patients in this study ranged from 18-69 years. Majority of the samples collected were from 40-49 (92) and the least were from 60-69 (1), patients within the age group 40-49 were mostly infected (11), none was infected from 60-69. Out of the 300 patient samples, 113 samples were from male patient, 107 samples from female patient in AE-FUTHA. While 40 samples were from male patients and 40 from female patients in Mater Hospital. From 133 samples in AE-FUTHA male had 12(5.4 %) positive samples and 8 (3.6 %) positive result in female, making a total of 20(9.0 %) positive samples from AE-FUTHA and 1 positive sample from a male patient in MMHA Afikpo, none was positive from their female counterpart.

Table 2: Demographic characteristics of critically ill patients with *A. baumannii* infections

Characteristics	AE-FETHA		MATER MISERICORDIAE	
	No of Sampled	No Positive (%)	No Sampled	No Positive (%)
Age (Yrs)				
18-29	31	3 (1.36)	22	0(0)
30-39	88	4 (1.81)	36	1(1.25)
40-49	94	11 (5.0)	19	0(0)
50-59	6	2 (0.90)	3	0(0)
60-69	1	0 (0)	0	0(0)
Sex				
Male				
Female	113	12(5.4)	40	1(3)
	107	8(36)	40	0(0)

at AE- FUTHA and Mater Misericordiae Hospital

Table 3, different wards where clinical sample were collected for this study include medical ward, surgical ward, orthopedic ward and ICU. From AE-FUTHA total number of samples collected from medical ward were 89, 15 (7.2 %) were positive, 65 were collected from orthopedic ward with 1 (0.45 %) positive, surgical ward had a total of 61 sample with 3(1.3 %) positive and 5 samples from ICU with only 1(0.45 %) positive sample. While from MMHA 29 clinical samples were collected from medical ward, 36 from orthopedic ward and 15 from surgical ward. Only 1(1.25 %) positive sample was from orthopedic ward, none was gotten from medical and surgical ward of the hospital.

Table 3: Frequency distribution of critically ill patients with *Acinetobacter baumannii* infection by the hospital wards/units at AE- FUTHA and Mater Misericordiae Hospital

Ward	AE-FUTHA		MATER	
	No Sampled	No Positive (%)	No Sampled	No Positive (%)
Medical ward	89	15(6.8)	29	0(0)
Orthopedic ward	65	1(0.45)	36	1(1.25)
ICU	5	1(0.4)	0(0)	0(0)
Surgical ward	61	3(1.3)	15	0(0)
Total	220	20(9.0)	80	1(1.25)

Frequency and percentage isolation of *A baumannii* pathogen from clinical sample

As shown in Table 4, the highest number of samples collected from AE-FUTHA was from Catheter urine (70), followed by wound sores (67), oral swab (26), wound drain (24), Nasal swab (20) and skin swab (13). From Mater Hospital catheter urine (50) was also the highest sample collected followed by wound sores (15), Nasal swab (5), wound drain (5), oral swab (3) and skin swab (2). Results showed that the most common part were *A. baumannii* pathogens were isolated from AE-FUTHA were from catheter urine (16%), wound sores (9%), and skin swab (8%), no *A. baumannii* pathogen was found from Nasal and oral swab. From Mater Hospital only (2%) *A. baumannii* pathogen was found from catheter urine and non from the other samples.

Table 4: Frequency distribution of critically ill patients with *Acinetobacter baumannii* infections with respect to clinical sample types at AE-FUTHA and MMHA

Sample source	AE-FUTHA		MATER HOSPITAL	
	TNSA	TNAB (%)	TNSA	TNAB (%)
Catheter urine	70	11(16)	50	1(2)
Wound sores	67	6(9)	15	0(0)
Skin swab	13	1(8)	2	0(0)
Nose swab	20	0(0)	5	0(0)
Mouth swab	26	0(0)	3	0(0)
Wound drain	24	2(8)	5	0(0)
Total	220	20(10)	80	(1%)

Keys: TNSA= Total Number of Samples Analyzed; TNABI= Total Number of *A. baumannii* Isolated.

Antibiotic Resistance Patterns of *A. baumannii* Isolated in Clinical Sample

Antimicrobial susceptibility test of the 21 *A. baumannii* clinical isolates was subjected to 10 commonly used antibiotics. *A. baumannii* strains showed the highest levels of resistance against Tetracycline (TE) (100 %), Trimethoprim sulphamethoxazole (SXT) (100 %), followed by Amikacin (80.9 %), Ceftriaxone (80.9 %), Gentamicin (CN) (57 %), Doxycycline 10(47.6 %), Ciprofloxacin (Cip) 9(42.8 %) while Meropenem, Imipenem and Polymyxin B had a low resistance rate of 3(14.2 %), 4(19.0 %) and 7(33.3 %) respectively (Table5).

Table 5: Antibiotics resistance patterns of *A. baumannii* isolated in clinical sample

Sample source	Disc strength	No RESISTANCE (%)	No SENSITIVE (%)
Ciprofloxacin (Cip)	(5 µg)	9(42.8)	12(57.1)
Tetracycline (TE)	(5 µg)	21(100)	0
Trimethoprim-sulphamethoxazole(SXT)	(25 µg)	21(100)	0
Imipenem (IPM)	(10 µg)	4(19.0)	17(80.9)
Gentamycin (CN)	(10 µg)	12(57.1)	9(42.8)
Amikacin	(30 µg)	17(80.9)	4(19.0)
Meropenem(MEM)	(10 µg)	3(14.2)	18(85.7)

Doxycycline		10(47.6)	11(52.3)
Polymyxin B	(300unit)	7(33.3)	14(66.6)
Ceftriaxone	(30 µg)	17(80.9)	4(19.0)

Key: = IMP = Imipenem (10 µg), TET=Tetracycline (10 µg), MEM =Meropenem (10 µg), (SXT)Sulfamethoxazole/trimethoprim (25 µg) AMK = Amikacin (30 µg), CIP = Ciprofloxacin (5 µg), Polymyxin B, (300unit), Ceftriaxone (30 µg), Doxycycline and (CN) = Gentamicin.

Multiple Antibiotic Resistant Index

The MARI of individual *A. baumannii* from the two different sources was calculated and total MARI was seen as 12.1 while average MARI was 0.57. Bacteria having MARI (>0.2) originate from a high-risk source of contamination where several antibiotics are used. MARI value of ≤0.2 indicates strain originated from sources where antibiotics are seldom or never used

Molecular characterization of the isolates

Genotypic identification by 16S rRNA sequencing result showed that the 15 isolates tested were *A. baumannii* as all the isolates showed clear bands at 1500 bp in the gel electrophoregram of the 16S rRNA gene amplification (Figure 2).

The 15 isolates were further re-confirmed and identified as *A. baumannii* by sequencing as compared with the Gene Bank and their respective accession numbers as obtained were deposited in the NCBI database. The percentage pair-wise identity numbers of the 15 *A. baumannii* isolates ranged from 100, 99.87, 99.93, 99.87, 100, 99.80, 99.90, 99.93, 99.93, 99.93, 98.87, 99.73, 99.87, 99.73, and 99.93 (%) (Table). Out of the 15 *A. baumannii* isolates identified by PCR, 9 (60 %) was characterized to have the presence of the *OXA-23* gene. Molecular analysis of the 21 MDR *A. baumannii* isolates showed that 9 out of 15 molecularly characterised isolates from both sources harboured *bla*_{OXA-23-like} genes in AE-Federal University teaching hospital and Mater hospital.

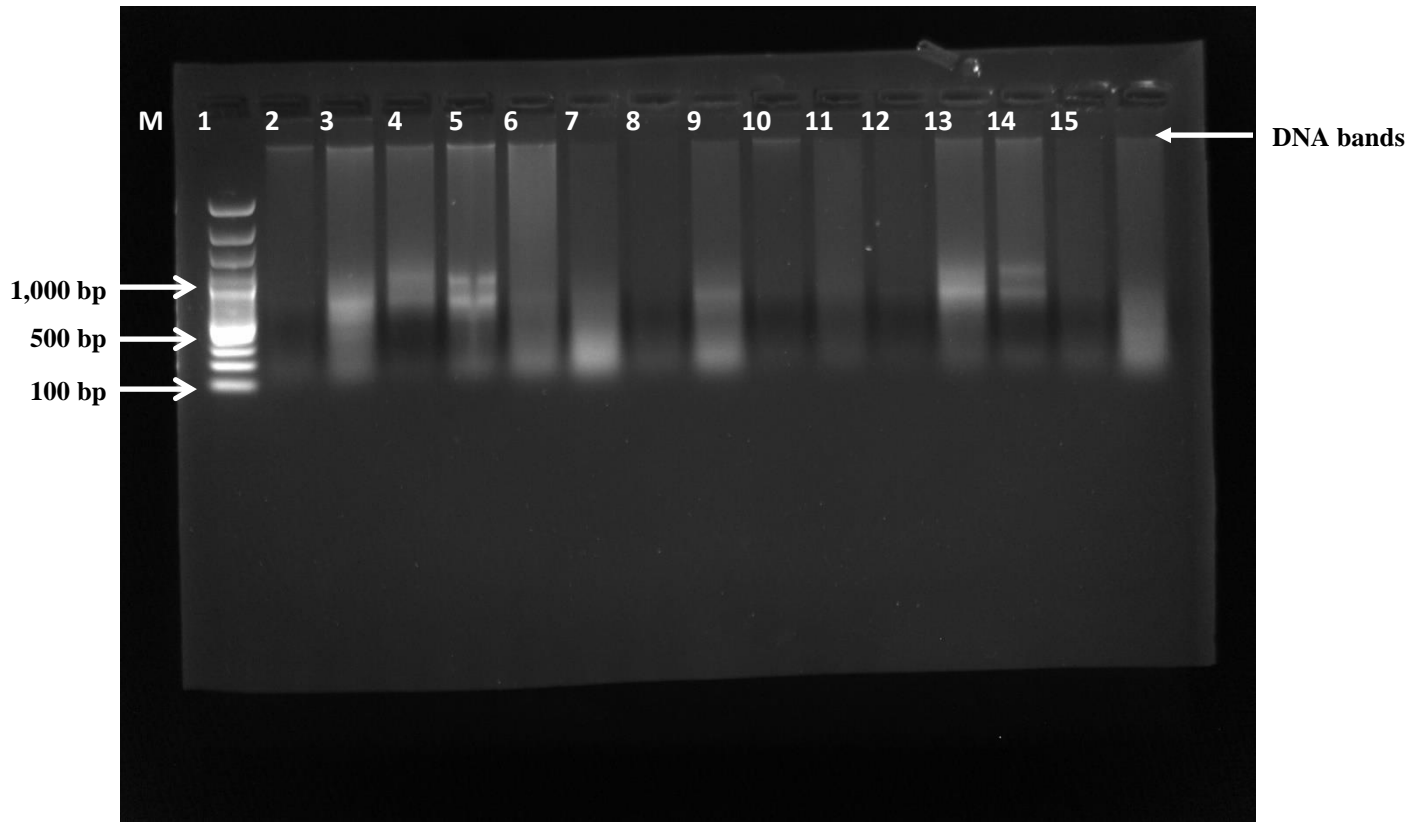


Fig 1: Integrity results showing the quality of DNA extracted from the fifteen *Acinetobacter baumannii* samples from Ebonyi State, Nigeria

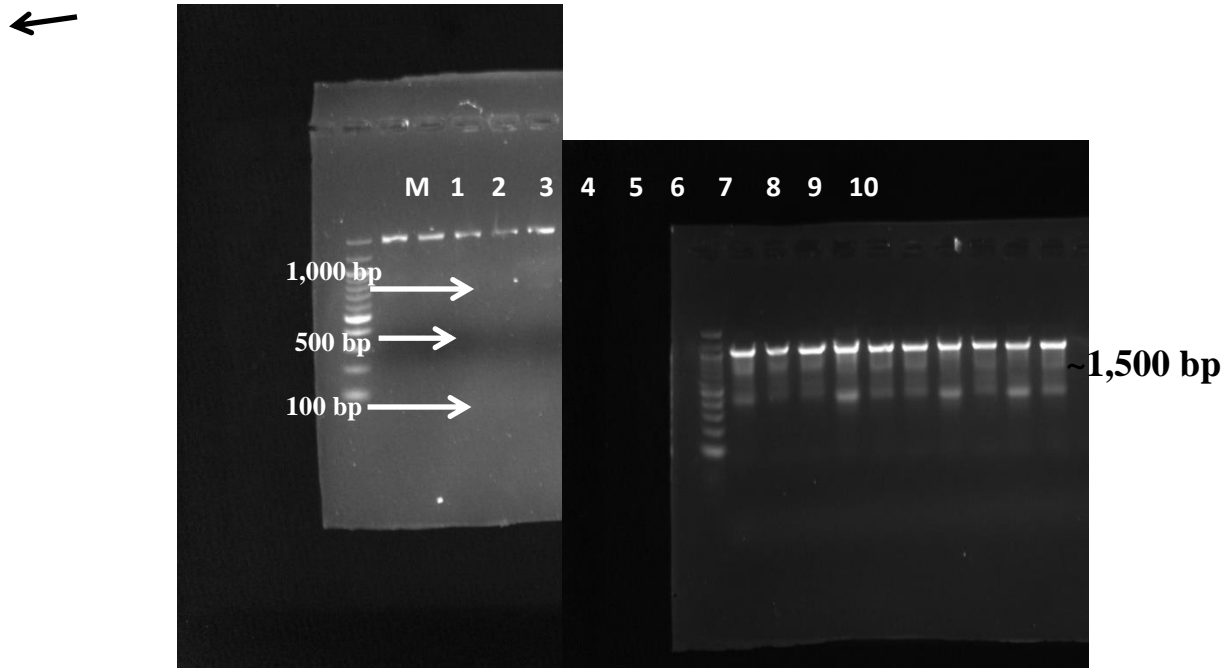


Fig 2: Polymerase chain amplification of the fifteen *Acinetobacter baumannii* samples from Ebonyi State, Nigeria using bacterial primers targeting the 16S ribosomal RNA segments at 1,500bp.

Table 10: Blast Result of of Partial 16S Ribosomal RNA Gene Sequences of the Bacterial Isolates from the two hospital

Sample ID	NCBI Accession Number	Properties(bp)	Pairwise Identity	E-value	Alignment score	Highest query Coverage (%)
AE01	OP985301	1,504	100.0 %	0.0	≥200	99
SW02	OP985302	1,501	99.87 %	0.0	≥200	100
ICU03	OP985303	1,502	99.93 %	0.0	≥200	100
ICU04	OP985304	1,502	99.87 %	0.0	≥200	99
SW05	OP985305	1,503	100.0 %	0.0	≥200	100
MW06	OP985306	1,502	99.80 %	0.0	≥200	100
MW07	OP985307	1,502	99.93 %	0.0	≥200	99
OW08	OP985308	1,501	99.93 %	0.0	≥200	100
MW09	OP985309	1,503	99.93 %	0.0	≥200	100
MW10	OP985310	1,503	99.87 %	0.0	≥200	99
MATER-11	OP985311	1,501	99.73 %	0.0	≥200	100
MW12	OP985312	1,503	99.87 %	0.0	≥200	100
MW13	OP985313	1,503	99.93 %	0.0	≥200	99
ICU14	OP985314	1,501	99.80 %	0.0	≥200	100
MW15	OP985315	1,502	99.93 %	0.0	≥200	99

The result of the phylogenetic relatedness among the isolates revealed that all the isolates are all *Acinetobacter baumannii* Figure 3.

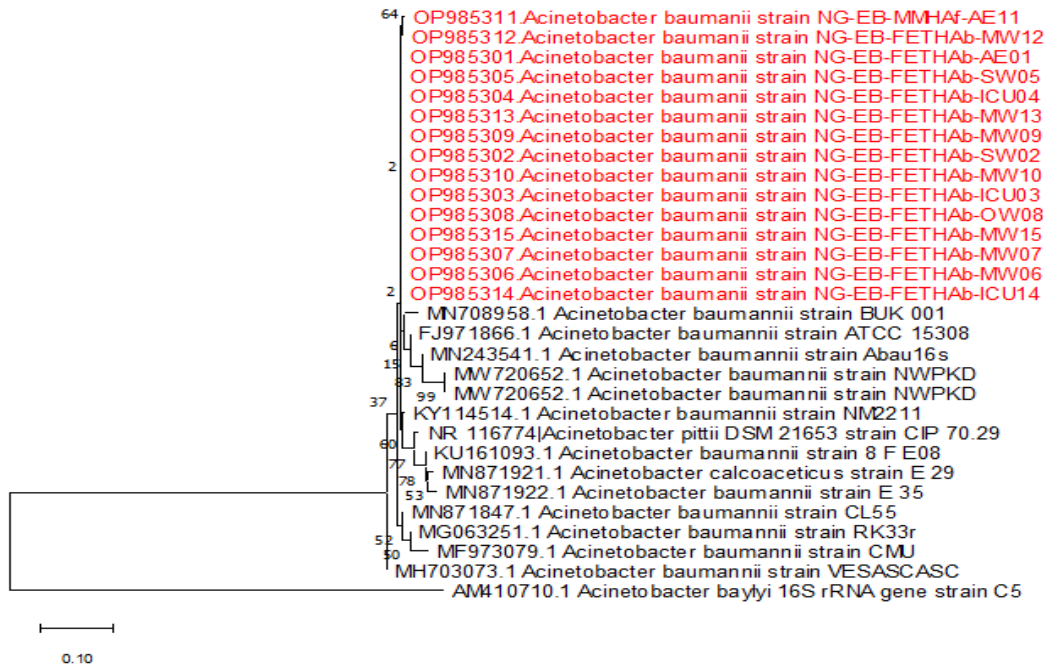


Figure 3: Phylogenetic tree showing evolutionary relationship of fifteen *Acinetobacter baumannii* samples obtained from Ebonyi State, Nigeria (in red) with other stains available in the GenBank database.

DISCUSSION

A. baumannii is a well-known opportunistic infection that mostly affects weakened persons, particularly those in intensive care units (ICUs) and those with pre-existing illnesses. This organism has consistently posed a threat to many medications (Egwu *et al.*, 2021). The incidence of *A. baumannii* in the investigations indicates that infections were more frequent in males, with a prevalence of 13 cases (8.5%), compared to females, with a prevalence of 8 cases (5.4%). The explanation may be attributed to the higher frequency of hospital visits among males compared to their female counterparts, with a difference of 8 (5.4%) percentage points. Egwu *et al.*, 2021 found that male patients (14, 6.9%) have a higher susceptibility to *A. baumannii* infection compared to female patients (9, 4.8%). According to Prashanth and Badrinath's (2006) study, the prevalence of infections was higher in males (58.00%) than in females (42.00%). This data indicates that males have a higher susceptibility to pathogenic organisms compared to females. The average age of patients from whom *A. baumannii* was isolated in this investigation ranged

from 18 to 59. The highest number of infections (11, or 5.0%) occurred in individuals between the ages of 40 and 49. According to Odewale *et al.* (2016), individuals between the ages of 41 and 70 are more prone to being infected by *A. baumannii*. According to Neetu *et al.* (2015), *Acinetobacter* infection was more prevalent among patients aged over 50 years. This indicates that *A. baumannii* infections are predominantly prevalent among patients aged 40 to over 50 years.

Our investigation revealed that the medical ward had the greatest count of *A. baumannii* isolates, with a total of 15, followed by the surgical ward with 4 isolates. *A. baumannii* is widely distributed in the environment and can be found in several locations within healthcare facilities. This study specifically identifies medical ward patients as being at a high risk of being colonised or infected by this disease. In contrast to Victor *et al.* (2014), who primarily focused on isolating patients from the ICU in Nigeria. Another study conducted by Natalia *et al.* (2018) found a higher prevalence rate of 18.4% among patients admitted to the surgical ICU at the University of Maryland Medical Centre in the USA. Neetu *et al.*, (2015) also verified that the rate of isolation and antibiotic resistance was greater in the Intensive Care Units (ICUs) of the hospital in Padmashree Bangalore. Our findings contradicted theirs but aligned with Egwu *et al.*, 2021, who reported the greatest number of *A. baumannii* positive samples in Nigeria from the Medical Ward.

In this study, a total of 21 isolates of *A. baumannii* were obtained from various clinical samples, including urine, nasal swab, wound drain, wound sores, skin swab, and oral swab. The findings revealed that the most prevalent locations for the *A. baumannii* pathogen were catheter urine (11 isolates, 16%), followed by wound sores (6 isolates, 9%) and skin swab (1 isolate, 8%). *A. baumannii* is a pathogen responsible for urinary tract infections (UTI), which are infections that affect the kidneys, ureters, or bladder. This can occur when a pathogen enters the body through the urinary tract. Additionally, it can infiltrate via a catheter employed for urinary drainage. (Natalia *et al.*, 2021).

The investigations conducted by Lone *et al.*, (2009) in Srinagar, India, Chakraborty *et al.*, (2011) in West Bengal, and Egwu *et al.*, 2021 in Ebonyi State, Nigeria have indicated that patients' urine samples and wound exudates/pus are the most common sites of *A. baumannii* pathogen. The investigation revealed that the isolated *A. baumannii* strains exhibited resistance to at least 3 classes of antibiotics, indicating that they were multidrug-resistant (MDR). The isolated *A. baumannii* exhibited the greatest resistance percentages to Tetracycline (100%), sulfamethoxazole/trimethoprim (100%), followed by Ceftriazone (80.9%), and Amikacin (80.9%). Additional antibiotics that are included in the list are gentamycin with a prevalence of 57.1%, doxycycline with a prevalence of 47.6%, and polymycin B with a prevalence of 33.3%. The medicines meropenem (85.7%) and imipenem (80.9%) were shown to be the most effective against *A. baumannii*. This finding aligns with the research conducted by Egwu *et al.* (2021), which concluded that meropenem and imipenem are the preferred medications for treating *Acinetobacter baumannii* infections. Our study findings align with those of Direkel *et al.*, 2016 and Eghbalimoghdam *et al.*, (2017) about the low resistance rate to imipenem in Turkey and Iran, respectively. These findings indicate that meropenem (85.7%) and imipenem (80.9%) are the most effective medications for treating *A. baumannii* in Ebonyi State, Nigeria. In a study conducted by Muhammed (2016), it was shown that *A. baumannii* exhibited a remarkably high resistance to gentamycin, with a resistance rate of 94.3%, and to tetracycline, with a resistance rate of 95%. Our results were consistent with theirs, but they differed from ceftriazone, which exhibited a lower resistance rate of 35% compared to our rate of 80%. The discrepancy may stem

from the fact that *A. baumannii* was isolated from various sorts of fluids, such as wound drains, whereas the bulk of our samples were derived from urine. The contrast may also be attributed to the improper utilisation of antibiotics. Al-Agamy *et al.* (2014) reported that all *Acinetobacter baumannii* isolates were found to be 85% resistant to Ciprofloxacin and 70% resistant to Imipenem. The present study by Al-Agamy *et al.* (2014) contradicts the findings of Al-Agamy *et al.* (2014), who saw a decline in resistance to both antibiotics. The study conducted Lone *et al.* (2009) found that the rates of antibiotic resistance for Ciprofloxacin, Imipenem, Ampicillin/sulbactam, Ceftazidime, and Amikacin were 100%, 99.4%, 99.4%, 99.4%, and 91.8%, respectively. The resistance to Ciprofloxacin, Ampicillin/sulbactam, Ceftazidime, and Amikacin seen in our investigation aligns with the findings of Lone *et al.* (2009). The MARI of *A. baumannii* from the two separate hospitals was determined. The findings from our research indicate a cumulative MARI of 12.1 and an average MARI of 0.57, which aligns with the findings of Yaw *et al.* (2019) where their average MARI exceeded 0.20. Bacteria with a minimum inhibitory concentration (MIC) of more than 0.2 originate from a source of contamination that poses a high danger, when many antibiotics are utilised. A MARI score of ≤ 0.2 suggests that the strain comes from sources where antibiotics are rarely or never utilised.

CONCLUSION

The organism's capacity to endure and establish itself in the hospital environment for an extended duration results in a significant infection that is commonly escalating within hospitals. Our analysis revealed that catheter urine and wound sores were the samples most prone to colonisation, and older patients had a higher susceptibility to *A. baumannii* infection compared to younger patients. In our study location, *A. baumannii* has displayed a significant prevalence of multidrug resistance. Meropenem, imipenem, and Polymyxin B have proven to be the most effective antibiotics for treating *A. baumannii* infections.

Conflict of Interest: The authors declared no conflict of interest

Funding source: This study was self-funded by the authors.

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