

Original article

Detection of Multidrug-Resistant *Acinetobacter baumannii* among Gram-Negative Bacteria Isolated from Clinical Samples

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Abstract

Acinetobacter baumannii is an aerobic, Gram -negative cocco-bacilli, non-fermentative, non-motile, and non-fastidious organism belonging to the genus Acinetobacter. The A. baumannii has emerged as a worldwide nosocomial pathogen causing about 80% of nosocomial infections comprising ventilator-acquired pneumonia, bacteremia, meningitis, urinary tract infections, skin and soft tissues infections associated with high mortality rate of approximately 63.3%. Although literature shows sufficient information about the drug resistant A. baumannii, there has been inadequate reports on the antibiotic resistance level of this bacterium in the study area. The aim of this research was to detect Multidrug-resistant A. baumannii isolates among Gram-negative bacteria isolated from Federal Teaching Hospital, Gombe, Nigeria. A total of 1008 clinical samples were collected and cultured on MacConkey agar and Blood agar plates at 370 C for 18-24 hours. Following the incubation period, discrete colonies obtained were subjected to Gram staining. The Gram-negative isolates were identified based on conventional biochemical tests with further use of VITEK 2 COMPACT (BioMérieux, France) for confirmation of A. baumannii amongst the Gram-negative organisms. The results obtained showed that 263 Gram-negative organisms were isolated. A. baumannii accounted for 8.5% prevalence. Most of the A. baumannii isolated were from the male patients (75%) within the age range of 33-48 years. Antibiotic susceptibility test using Kirby Bauer method in accordance with CLSI guidelines was done on 20 A. baumannii isolates. The isolates were more sensitive to levofloxacin (60%), followed by Gentamicin (55%), then Ciprofloxacin and Tetracycline (50%) respectively. High level of resistance to Ceftriaxone (80%), Cefepime (75%), Ceftazidime (65%), Piperacillin-Tazobactam (55%), Ampicillin/Sulbactam (60%), Tigecycline (60%), Meropenem (55%) and Amikacin (60%). This study revealed that 15 (75%) of the A. baumannii were found to be multidrugresistant. Therefore, antibiotic stewardship is necessary to combat further dissemination of this organism.

Keywords: Gram-negative bacteria, Acinetobacter species, nosocomial infections, susceptibility test, multi-drug resistance.

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INTRODUCTION

Acinetobacter baumannii is an important opportunistic pathogen causing about 80% of clinical infections of the 21st century (Odewale, Adefioye, Ojo, Adewumi, & Olowe, 2016; Almasaudi, 2018; Khaledi et al., 2019), and several nosocomial infections such as ventilator-associated infection (VAP), burn and wound infections leading to significant morbidity and mortality particularly in immuno-compromised patients as well as those admitted in intensive care units (Basri et al., 2015; Azimi, Talebi, Pourshafie, Owlia, & Lari, 2015; Asif, Alvi, & Rehman, 2018). Report showed that, the *A. baumannii* has been isolated from blood, sputum, skin, pleural fluid, and urine samples, usually in device associated infections (Mayasari & Siregar, 2015). This organism could survive for extended periods in the environment due to its ability to form biofilms, a bacterial community enclosed in a matrix of self-produced extracellular polysaccharides that enable attachment to hospital medical equipment and surfaces. In fact, infections caused by biofilms producing organisms are comparatively more difficult to treat (Ibrahim et al, 2019).

The *A. baumannii* is an aerobic, Gram -negative cocco-bacilli, non-fermentative, non-motile, and non-fastidious organism belonging to the genus Acinetobacter, family Moraxellaceae and order Pseudomonadales (Nemec et al, 2016). This bacterium is oxidase negative, urease negative, citrate positive, indole negative and catalase positive with optimal growth temperature of 37 °C. However, some environmental isolates are capable of growing in a temperature range of 20 °C -30 °C (Asif et al., 2018; Almasaudi, 2018; Shareef and Risan, 2021).

The *A. baumannii* has been involved in blood stream infections and accounts for about 15% cases due to invasive procedures such as intravascular or respiratory catheters, endotracheal tubes or cannulas (Mirnejad, Moradli, Mirkalantari, & Golmohammadi, 2018). This can lead to prolonged hospitalization due to treatment failure (>14 days) which increases the treatment cost (Bashir et al., 2019). The *A. baumannii* has been described by Al-Hassan and Al- Madboly (2020), as an important globally distributed hospital acquired Gram-negative pathogen that has emerged with a great tendency to causing outbreaks, especially amongst patients admitted in the intensive care units. Over the past 30 years, the pathogen has proven difficulty of being controlled by the conventional antimicrobials provided for the healthcare services, as such regarded as a dreadful lethal agent with a genome set up for prompt development of resistance to most of the available antimicrobial agents (Rizk, Elwakil, & Attia, 2021)).

Worldwide, antimicrobial resistance has been a major clinical problem and a threat to healthcare system. Microbial resistance arises due to development of various resistance mechanisms like drugs modification and their inaccessibility to microbial target sites (Ferri, Ranucci, Romagnoli, & Giaccone, 2017; Garba et al., 2021a). Escalation of resistance to antimicrobial agents amongst bacterial pathogens is quite worrisome and could be majorly as a result of adaptable microbial genetic system under the pressure of diverse control agent(s) (Garba et al., 2018).

Antimicrobial resistance has been associated to inappropriate and overuse of therapeutic agents coupled to lack of development of novel drugs by the pharmaceutical industries, perhaps as a result of regulatory challenges and economic reasons. Centre for Disease Control and Prevention (CDC) shows that many bacteria exhibit serious threat to effective clinical treatments, increase financial cost on government healthcare systems, and patients due to antimicrobial resistance (Garba et al., 2021b).

The A. baumannii has been counted amongst six (6) significant MDR microorganisms occurring in clinical settings worldwide. The rates of A. baumannii resistance differ from one location to another which increases over time. High resistance rates of this organisms to various antimicrobial agents have been reported. For instance, its resistance rates are known to antibiotics like gentamycin and ceftazidime (0-81%), ciprofloxacin (19 -81%), amikacin (10 -51%), and piperacillin-tazobactam (36 to 75%) (Al-Tamimi et al., 2022). Imipenem and meropenem used to be the most effective drugs against the A. baumannii infections. Nevertheless, recent reports confirmed 87% resistance to these drugs by the A. baumannii based on clinical setting and geographic distribution which leaves colistin/tigecycline being the only available treatment options for MDR A. baumannii infections. Unluckily, colistin and/or tigecycline have also been resisted in Europe giving rise to 3–6% incidence rate. It is quite worrisome to note that Acinetobacter species have developed resistance to virtually every available antimicrobial agent (Al-Tamimi et al., 2022). Although the prevalence (8.5%) and antibiotic resistance profile of A. baumannii have been reported from Ladoke Akintola University Teaching Hospital, the southern part of Nigeria (Odewale et al., 2016), to date there is limited data on nosocomial MDR A. baumannii in the North-East to the best of our knowledge. The aim of this study was to investigate multidrug-resistance pattern of A. baumannii from Federal Teaching Hospital Gombe.

MATERIALS AND METHODS

Study setting

The research was conducted at Federal Teaching Hospital, Gombe (FTHG) from January, 2022 to April, 2022. The FTHG is located within the city of Gombe in the North-Eastern part of the Federal Republic of Nigeria, on latitude 10.283333, and longitude 11.166667 with an estimated population of 261,536. This hospital is 800-bed capacity established in 1996 by the Federal Government. There are eleven [11] main wards in the hospital including the amenity ward. The various clinical departments run specialist and sub-specialist clinics from Monday to Friday. Emergencies are attended to on a 24 -hour basis through the Accident & Emergency Unit [A & E]. The hospital is strategically located for access to both rural and urban population throughout the state.

Ethical Approval

This research was approved by the Research and Ethics Committee, Federal Teaching Hospital, Gombe, Nigeria with a reference number of **NHREC/25/10/2013**.

Sample Collection

A total of 1008 clinical samples from various clinics of Federal Teaching Hospital, Gombe were collected for this study. The samples were comprised of respiratory tract specimens (110), pus and swabs (141), CSF and other sterile body fluids (55), urine (628), catheter tips/urethral specimens (6) and blood (68) submitted to Medical Microbiology/Immunology laboratory of the hospital within period of four (4) months (January to April, 2022) were processed according standard procedures (Cheesbrough, 2010).

Isolation and identification of Gram-negative bacteria

Blood agar base and MacConkey agar plates ((Sigma-Aldrich) used to isolate the bacteria were prepared according to manufacturer's instructions. All samples were cultured on these media and incubated at 37 °C for 18-24 hours. Following the overnight incubation, bacterial identification was based upon cultural characteristics, Gram staining and biochemical tests. Further confirmation of *Acinetobacter* species was done using VITEK 2 ID-GNB (BioMérieux, France) assay according to manufacturer's recommendation (Al-Tamimi, *et al.*, 2022; Mahmood & Al-Berfkani, 2022). Detailed description of identification procedures has been given under the subheadings below.

Gram staining

A single smear was made from each bacterial colony and emulsified in a drop of 0.9% normal saline on a grease free glass slide, allowed to air dry and fixed by passing over Bunsen flame three times. Crystal violet stain (Sigma-Aldrich) was added to the smear, left to stand for 1 minute and washed off the stain with running tap water. Lugol's iodine (Sigma-Aldrich) was added for 1 minute, then washed with running tap water and decolorized with acetone (Sigma-Aldrich) for 20 seconds. The smear was rinsed with tap water and counterstained with safranin solution (Sigma-Aldrich) for 1 minute. The smear was washed with running tap water and allowed to air dry (Cheesbrough, 2010; Garba et al, 2021). The smear was examined microscopically using X100 oil immersion objective lens of a light microscope (Leica DM 500).

Biochemical tests

The following biochemical tests have been carried out to conventionally identify the Gramnegative isolates according to Cheesbrough, 2010.

Catalase test

A single colony of each isolate from an overnight culture on nutrient agar (Sigma-Aldrich) plate was emulsified in a drop of normal saline on microscopic slides. Two drops of 3% hydrogen peroxide (H₂O₂,) (Sigma-Aldrich) were added and observed immediately for effervescence.

Citrate utilization test

Simmon citrate agar (Oxoid, England) was prepared in bijou bottles according to manufacturer's instructions. A sterile straight wire loop was used to pick up a pure colony of the test organism, stabbed butt of the medium and streaked the slope. The bottles were incubated at 37 $^{\circ}$ C for 18 -24 hours under aerobic conditions.

Indole test

The test organism from an overnight culture on Mueller Hinton agar (Sigma-Aldrich, USA) was inoculated in a bijou bottle containing 3 ml of sterile peptone water and incubated at 37 °C for 18-24 hours. After the incubation, 0.5 ml of Kovac's reagent (Fisher Scientific) was added, shaken gently and examined within 10 minutes.

Oxidase test

Two (2) drops of freshly prepared oxidase reagent was dropped onto a piece of Whatman filter paper. A single colony from an overnight culture on Mueller Hinton agar was picked with a glass rod to make smear of the bacterial colonies on the spot of oxidase reagent and observed within 10 seconds.

Motility test

A semi solid agar medium which contained 0.2-0.4% agar in 10 ml peptone water was prepared in a test tube. A sterilized straight wire loop was used to pick up the test organism and stabbed the medium followed by incubation at 37 °C for 18-24hours.

Triple sugar iron (TSI) agar test

This medium was prepared in test tubes according to the manufacturer's instructions. A sterilized straight wire loop was used to pick up a colony of each test organism from an overnight culture on Mueller Hinton agar. The butt of the tube was stabbed and slant streaked. Incubation was done at 37 °C for 18-24hours.

Urease test

The Urea agar slant medium was prepared following manufacturer's instructions. A pure colony of each test isolate was picked up using a sterilized straight wire from an overnight culture on Mueller Hinton agar. The slope of the urea agar was streaked and the butt was stabbed. This was incubated at 37°C for 18-24 hours.

VITEK 2 COMPACT identification kit

This test was performed to confirm the suspected colonies of *Acinetobacter baumannii* amongst the Gram-negative bacteria. A colony from an overnight culture of Acinetobacter species on MacConkey agar was picked up and prepared its suspension in 3 mL of normal saline that matched 0.5 McFarland Standard using DENSICHEK® Plus (BioMérieux Inc DensiCHEK[™], France). The GN:cards (BioMérieux, France) and cassette barcodes of the kit were scanned to establish traceability. The cards inoculated with the standardized bacterial inoculum were placed inside the cassette and loaded inside the VITEK 2 COMPACT machine for verification. After verification of the code it was manually transferred from filling door to loading door for automated processing within 5-8 hours. The VITEK 2 system used authenticated all the Acinetobacter species as described by the manufacturer (BioMérieux, France).

Detection of Multidrug-resistant Acinetobacter baumannii

Detection of multidrug-resistant *A. baumannii* was achieved by testing the organism against seven (7) different classes of antibiotics based on CLSI guidelines as demonstrated under the following subheadings:

Inoculum standardization

A sterile wire loop was used to pick up a few well-isolated colonies of the *A. baumannii* from an overnight culture on Mueller Hinton agar and emulsified in 4 ml of sterile physiological saline. Turbidity of the suspension was matched to 0.5 McFarland Standard (Cheesbrough, 2010; Duruike, Affia, Nyenke, & Konne, 2022) and confirmed by the McFarland Standard DENSICHEK® plus of the VITEK 2 system (Marcy L'Etoile, France).

Antibiotic Sensitivity Test

The sensitivity test was determined by using modified Kirby-Bauer disk diffusion method on Mueller –Hinton agar (Oxoid, Cambridge UK) following standard procedures recommended by the Clinical and Laboratory Standard Institute Wayne, USA (CLSI, 2021).

Precisely, a sterile swab stick was used to inoculate a plate of Mueller Hinton agar with the standardized inoculum of the test organism after removing excess fluid by pressing and rotating the stick against the side of the tube above the level of the suspension. Then, the swab was streaked evenly over the surface of the Mueller-Hinton agar plate in three directions, rotating the plate approximately 60° to ensure even distribution. After streaking, the plate was allowed to stand for 3-5 minutes for the surface of the agar to dry. A sterile forcep was used to place antibiotic discs about 15 mm from the edge of the plate and no closer than 25 mm from disc to disc (Cheesbrough, 2010). Twelve (12) antibiotics tested included ampicillin-sulbactam (10/10 µg), ceftazidime (30 µg), gentamicin (10 µg), ciprofloxacin (5 µg), neropenem (10 µg), piperacillin-tazobactam (100/10 µg), Ceftriaxone (30 µg), Tigecycline (10 µg), cefepime (30 µg), tetracycline (30 µg), amikacin (30 µg), from Oxoid Laboratories. All plates were incubated at 35 °C for 24 hours, after which diameter of inhibition zone of each of the antibiotic disk was determined by measuring size of clear zones with a graduated ruler. The measurement was done in millimetres and the zones were compared with the CLSI standards for interpretation (CLSI, 2021).

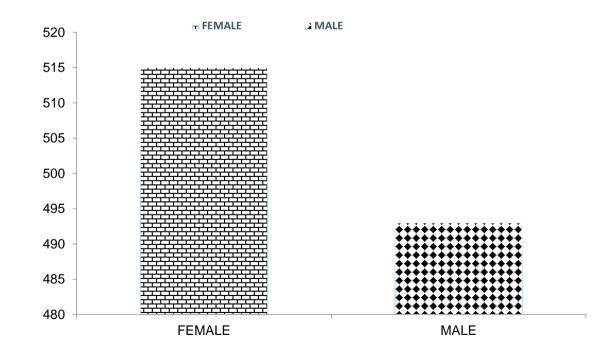
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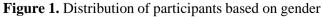
Demographic information

During the study period, a total of one thousand and eight (1008) participants from different age groups and gender were enrolled. Summary of the age distribution is shown in Table 1 with the highest participants within 33-48 years (31.3%), followed by 16-32 years (29.8%),<15years (18.8%), 49-64 years (12.9%) while the remaining 7.0% of the participants are >65 years of age. According to gender, the samples collected and analyzed included 515 (51.1%) females and 493 (48.9%) male subjects (Figure. 1).

Age Groups (years)	Frequency (n)	Percentage (%)
≤15	190	18.85
16-32	301	29.86
33-48	315	31.25
49-64	131	13.00
≥65	71	7.04
Total	1008	100.00

Table 1. Distribution of study participants based on age groups





Isolation and identification of bacterial Isolates

Following an overnight incubation of all samples on blood agar base and MacConkey agar, cultural characteristics showed 155(54.77%) and 128(45.23%) colonies to be lactose and non-lactose fermenting organisms, respectively. The Gram staining reactions confirmed the colonies as Grampositive and Gram-negative organisms as depicted in Figure 2. The Gram-negative bacteria had the highest number of occurrences with 283 isolates compared to the Gram-positive ones with only 172 isolates as shown in Table 2.

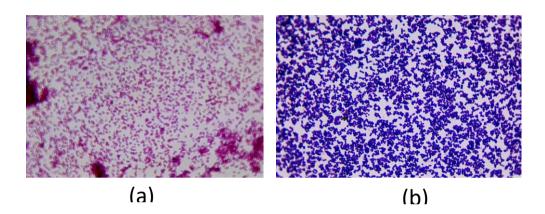


Figure 2. Microscopic examination of Gram Negative (a) and Gram positive (b) bacteria

Gram Reaction	Occurrence	Percentage (%)
Gram-negative	283	62.2
Gram-positive	172	37.8
Total	455	100

Table 2. Gram reaction of bacterial isolates

The Gram-negative isolates that have been subjected to various biochemical tests were successfully identified as *Proteus* spp (comprising *Proteus vulgaris* and *Proteus mirabilis*), *Providencia* spp, *Salmonella* spp, *Serratia marcescens*, *Shigella* spp, *Yersinia pestis*, *Burkholderia cepacia*, *Acinetobacter* species, *Alcaligenes faecalis*, *Pseudomonas aeruginosa*, *Rhizobium* radiobacter, *Citrobacter freundii*, *Escherichia coli*, *Enterobacter* aerogenes, and *Klebsiella* species (Table 3).

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Bacterial isolates	Biochemical test									
	Glu	Lact	Suc	H_2S	Gas	Cit	Urea	Ind	Mot	Ox
Proteus spp	+	-	-	+	+	+	+	+/-	+	-
Providencia spp	+	-	-	-	-	+	+	+	+	-
Salmonella spp	+	-	-	+	+	-	-	-	+	-
Serratia marcescens	+	-	-	-	-	+	-	-	+	-
Shigella spp	+	-	-	-	-	-	-	+	-	-
Yersinia pestis	+	-	-	-	-	-	+	+	+	-
Burkholderia cepacia	+	_	+	-	+	+	+	-	+	-
Acinetobacter species	-	_	_	_	_	+	_	_	+	_
Alcaligenes faecalis	-	+	_	_	_	+	_	_	+	+
Pseudomonas aeruginosa	-	-	_	_	_	+	_	_	+	+
Rhizobium radiobacter	+	_	_	_	_	_	+	_	_	_
Citrobacter freundii	+	+	+	_	_	+	_	_	+	_
Escherichia coli	+	+	+	_	+	_	_	+	+	_
Enterobacter aerogenes	+	+	+	_	+	+	_	_	+	_
Klebsiella spp	+	+	+	-	+	+	+	+/-	_	_

 Table 3. Biochemical characterization of Gram-negative isolates

 $Key: Lact=Lactose \ fermentation, \ Suc=Sucrose \ fermentation, \ Glu=Glucose \ fermentation, \ Cit=Citrate \ utilization \ test, \ Mot=Motility, \ Ind \ =Indole \ test, \ Urea=Urease \ test, \ H_2S=Hydrogen \ sulphides \ production, \ Gas \ production.$

Occurrence of Acinetobacter species and other Gram- negative bacteria

Table 4 shows the percentage occurrence of bacteria identified from the clinical samples. Of the 283 Gram-negative bacteria identified, Escherichia coli has the highest number of occurrence corresponding to 80(28.30%), followed by Klebsiella species with 64(22.61), Pseudomonas aeruginosa with 59(20.84%), Acinetobacter species with 24(8.50%), Proteus species with 18(6.36%), Enterobacter aerogenes with 7(2.47%), Shigella species with 6(2.12%), Citrobacter freundii and Serratia marcenscens with 5(1.77%) each, then Salmonella species, Yersinia pestis, and Providencia species each with 4(1.31%). The least percentage occurrence was observed with Alcaligenes faecalis, Burkholderia cepacia and Rhizobium radiobacter each with 1(0.35%).

S/No	Isolates	Number	Percentage (%)
1	Acinetobacter species	24	8.50
2	Alcaligenes faecalis	1	0.35
3	Burkholderia cepacia.	1	0.35
4	Proteus spp.	18	6.36
5	Providencia spp.	4	1.31
6	Pseudomonas aeruginosa.	59	20.84
7	Rhizobium radiobacter	1	0.35
8	Salmonella spp.	4	1.31
9	Serratia marcescens.	5	1.77
10	Shigella spp.	6	2.12
11	Yersinia pestis	4	1.31
12	Citrobacter freundii	5	1.77
13	Escherichia coli	80	28.30
14	Enterobacter aerogenes	7	2.47
15	Klebsiella species	64	22.61
	Total	283	100.00

Table 4. Percentage occurrence of Gram- negative bacteria

Furthermore, the 24 isolates of *Acinetobacter* species were further subjected to VITEK 2 COMPACT system of identification, which differentiated them into three different species as *Acinetobacter baumannii*, *Acinetobacter iwoffii* and *Acinetobacter calcoaceticus*. The *A. baumannii* was found to be the most occurring with 20(83.3%), followed by *A. iwoffii* and *A. calcoaceticus* each with 2(8.3%) isolates (Table 5).

Table 5. Percentage occurrence of A. baumannii and other Acinetobacter species

Acinetobacter species	Occurrence (%)	
A. baumannii	20 (83.3)	
A. iwoffii	2 (8.3)	
A.calcoaceticus	2 (8.3)	
Total	24 (100)	

Antibiotic susceptibility pattern of A. baumannii

The antibiotic susceptibility profile of the *A. baumannii* is presented in Table 6. Of the twenty (20) isolates of the *A. baumannii* subjected to antibiotic susceptibility test, only 5(25%) of the isolates were found to be susceptible to the drugs while the remaining fifteen (15) isolates corresponding to 75.0% were found to be resistant to more than three classes of antibiotics. The isolates were more sensitive to Levofloxacin (60%) followed by Gentamicin (55%), then Ciprofloxacin (50%), and Tetracycline (50%). However, the bacterium demonstrated a high resistance level to Ceftriaxone (80%),

Cefepime (75%), Ceftazidime (65%), Tazobactam (55%), Ampicillin-Sulbactam (60%), Meropenem (55%), and Amikacin (60%). An isolate is termed multidrug-resistant if it is resistant to at least three classes of antimicrobial agents—all Penicillins and Cephalosporins (including inhibitor combinations), Fluroquinolones, and Aminoglycosides. Therefore, fifteen (15) isolates of A. baumannii, were observed as multidrug-resistant (Figure 3).

S/No	Antibiotics (µg)	No Sensitive (%)	No Resistant (%)
1	CN	11(55)	9(45)
2	CIP	10(50)	10(50)
3	CRO	4(20)	16(80)
4	FEP	5(25)	15(75)
5	TET	10(50)	10(50)
6	CAZ	7(35)	13(65)
7	LEV	12(60)	8(40)
8	TZP	9(45)	11(55)
9	SAM	8(40)	12(60)
10	MEM	9(45)	11(55)
11	AK	8(40)	12(60)
12	TGC	8(40)	12(60)

Table 6. The antibiotic susceptibility patterns of A. baumannii

Key:CN:Gentamicin,CIP:Ciprofloaxin,,CRO:Cetriaxone,LEV:Levofloxacin,TZP: Piperacillin-Tazobactam, SAM: Ampicillin-Sulbactam, MEM: Meropenem, AK: Amikacin, TGC: Tigecycline,

FEP: Cefepime, TET: Tetracycline, CAZ: Ceftazidime,

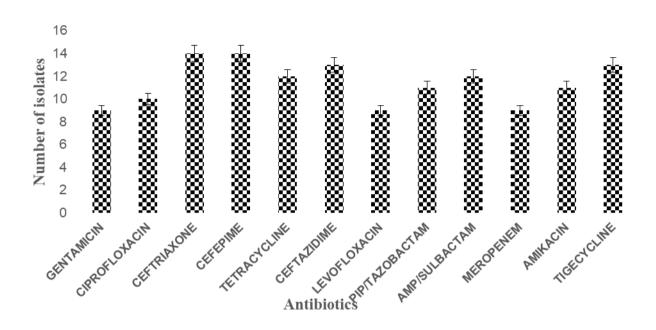


Figure 3. Pattern of Multidrug-Resistance of A. baumannii

Discussion

Occurrence of multidrug-resistant *Acinetobacter baumannii* in nosocomial infection has remained a great concern. The ability of this bacterium to cause infections has been attributed to different factors such as its ability to colonize human, especially healthcare workers and its survival in the environment (Duruike et al., 2022). This study evaluated the occurrence and antibiogram of *A. baumannii* isolated from Federal Teaching Hospital, Gombe, Gombe State, Nigeria. Detection of this organism in the clinical samples correlates well with earlier report which showed that the organism occurs in hospitalized patients (Duruike et al., 2022; Poorzargar, Javadpour, & Karmostaji, 2017)). Globally, this pathogen is of global public health concern as it poses a menace to both clinical and public health especially patients who are on therapeutic antibiotics (Duruike et al., 2022).

Of the 1008 clinical samples analyzed, 455 (45.1%) yielded positive isolates comprising 172 (37.8%) Gram-positive and 283 (62.2%) Gram-negative organisms. This is almost similar to the findings of Jain et al. (2019), who reported that 51% of clinical samples yielded positive results. Contrary to this study, Bashir et al. (2021) recorded 34.4% bacterial isolates. This could be attributed to differences in sample size and study region. Amongst the Gram-negative organisms isolated. *Acinetobacter* species was particularly the most significant pathogen, accounting for 8.5% of the nosocomial infections. The

increased occurrence of Gram-negative bacteria, including *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and Enterobacteriaceae in severe healthcare-associated infections has evolved as a significant clinical threat for medical fraternity in the recent decades (Parajuli et al., 2017).

Abdullah and Merza (2019) reported 6.8% prevalence of *A. baumannii* in Iraq, which was very close to the prevalence (8.5%) obtained in this study. However, Bashir et al., 2021 reported a slightly higher prevalence (10.1%) compared to that obtained in this report. Contrarily, the prevalence (0.7%) of *A. baumannii* reported by Heydarpour, Rahmani, Heydarpour, & Asadmobini, (2017) in Iran was lower than that obtained in this study. These differences could be attributed to different geographical locations and climate (Abdullah and Merza, 2019).

Antibiotic sensitivity test showed most of the *A. baumannii* isolates were resistant to commonly prescribed antibiotics such as Ceftazidime (65%), Ceftriaxone (80%), Cefepime (75%), among the Cephalosporin and Ampicillin–sulbactam (60%), Tigecycline(60%) and Amikacin(60%) among the β -lactam drugs. This may be the response to selective pressure since these later drugs are the most commonly used Cephalosporin in the hospital setting where this study was carried out. A resistance level of Ceftriaxone (85.7%) was reported by Almaghrabi, Joseph, Assiry, & Hamid (2018), which is almost similar to this present study. Also, in another study by Joshi *et al*, 2017, high resistance level to the Cephalosporin (Ceftazidime, Ceftriaxone, Cefotaxime) was reported rendering these drugs ineffective against the *A. baumannii*.

The global prevalence of nosocomial infections amounts to 1.40 million patients, which correspond to approximately 9% (Tarigan, Nawan & Toemon, 2023). The WHO report revealed that fifty five (55) or 8.70% of hospitals from different countries of Europe, Southeast Asia, Middle East and Pacific are burdened by nosocomial infections (Hapsari, Wahyuni & Mudjianto, 2018). These infections remain the serious problem and amongst the most frequent causes of morbidity and mortality in healthcare settings. The nosocomial infections may be as a result of endogenous (e.g., patients' normal flora) or exogenous (via contaminated objects or devices) factors within the hospital (Gill, Singh, Thapliyal & Karpoormath, 2019). Literature shows that the most commonly reported nosocomial pathogens are Staphylococcus *aureus, Pseudomonas aeruginosa, Escherichia coli, Klebsiella pneumoniae* and *Enterobacter* spp (Vaiyapuri, Joseph, Rao, Lalitha & Prasad, 2019).

In addition to *A. baumannii* and other Gram-negative bacteria, several Gram-positive bacteria have been reported to cause various nosocomial infections. For instance, Yang et al., 2023 reported high prevalence of *Staphylococcus aureus* among Gram-negative nosocomial bacteria. Compared to aerobic Gram-negative bacteria which account for approximately 60% of nosocomial infections, Gram-positive bacteria are associated with only 30% of hospital acquired infections. Moreover, 7% of nosocomial illnesses are caused by viruses and fungi (Saba and Balwan, 2023). Furthermore, both Gram-positive

and Gram-negative bacteria were detected in nosocomial samples in a study conducted by Nimer (2022) with corresponding prevalence of 43% and 57%, respectively.

Conclusion

The result of this finding showed the prevalence of *A. baumannii* among the Gram-negative organisms frequently isolated in Federal Teaching Hospital, Gombe and its antibiotic susceptibility profile. Three different species of *Acinetobacter* were found to be associated with hospital associated infections (*A, baumannii*, *A. iwoffi* and *A. calcoaceticus*). The most common was *A. baumannii* which was found in all major units of the hospital and showed MDR patterns. This study forms the baseline for the prevalence of *A. baumannii* in the hospital. Proper patients' management and antibiotic stewardship is recommended to reduce the spread of nosocomial infections due to Multidrug-resistant *A. baumannii*. Moreover, further study to identify antibiotic resistance genes in the *A. baumannii* with a view to reducing treatment failure and cost of nosocomial infections is recommended.

Conflict of Interest

The authors declare that they have no any conflict of interest.

REFERENCES

- Abdullah, Z. H., & Merza, N. S. (2019). Phenotypic and Molecular detection of *Acinetobacter baumannii* isolated from patients in Duhok city-Iraq. *Science Journal of University of Zakho*, 7(4), 132-137.
- Al-Hassan, L. L., & Al-Madboly, L. A. (2020). Molecular characterisation of an *Acinetobacter baumannii* outbreak. *Infection Prevention in Practice*, 2(2), 100040.
- Almaghrabi, M. K., Joseph, M. R., Assiry, M. M., & Hamid, M. E. (2018). Multidrug-resistant Acinetobacter baumannii: An emerging health threat in aseer region, Kingdom of Saudi Arabia. Canadian Journal of Infectious Diseases and Medical Microbiology, 2018.
- Almasaudi, S. B. (2018). *Acinetobacter* spp. as nosocomial pathogens: Epidemiology and resistance features. *Saudi journal of biological sciences*, 25(3), 586-596.
- Al-Tamimi, M., Albalawi, H., Alkhawaldeh, M., Alazzam, A., Ramadan, H., Altalalwah, M., & Hijawi, K. (2022). Multidrug-Resistant *Acinetobacter baumannii* in Jordan. *Microorganisms*, *10*(5), 849.
- Asif, M., Alvi, I. A., & Rehman, S. U. (2018). Insight into *Acinetobacter baumannii*: pathogenesis, global resistance, mechanisms of resistance, treatment options, and alternative modalities. *Infection and drug resistance*, *11*, 1249.
- Azimi, L., Talebi, M., Pourshafie, M. R., Owlia, P., & Lari, A. R. (2015). Characterization of carbapenemases in extensively drug resistance *Acinetobacter baumannii* in a burn care center in Iran. *International journal of molecular and cellular medicine*, 4(1), 46.
- Bashir, A., Aliero, A. A., Idris, A. M., Takalmawa, H. U., Faruk, S., & Agwu, E. (2019). Molecular characterization of *Acinetobacter baumannii* from patients with prolonged hospital stays in three

tertiary hospitals of Kano Metropolis, Northwestern Nigeria. African Journal of Microbiology Research: 13(27); 510-517

- Basri, R., Zueter, A. R., Mohamed, Z., Alam, M. K., NORSA'ADAH, B. A. C. H. O. K., Hasan, S. A., Ahmad, F. (2015). Burden of bacterial meningitis: a retrospective review on laboratory parameters and factors associated with death in meningitis, Kelantan Malaysia. *Nagoya journal of medical science*, 77(1-2), 59.
- Cheesbrough M. (2010). District Laboratory Practice in Tropical Countries. Cambridge University
- Duruike, P. N., Affia, A., Nyenke, C. U., & Konne, F. E. (2022). Multi Drug Resistance and Multi Antibiotics Resistance Index of Acinetobacter baumannii Isolated from Hospitals in Port Harcourt Metropolis. *Microbiology Research Journal International*, 1-11.
- Ferri, M., Ranucci, E., Romagnoli, P., & Giaccone, V. (2017). Antimicrobial resistance: A global emerging threat to public health systems. *Critical reviews in food science and nutrition*, *57*(13), 2857-2876.
- Garba, L., Abdulfatah, A., Adamu, M. T., Ismail, H., Yusuf, I., & Dahiru, A. S. (2021a). Evaluation of Antibacterial Potency of Endophytic Fungi Isolated from Mentha piperita.
- Garba, L., Muhammad, M. B., Adamu, M. T., Isa, S., Abdullahi, M. M., & Yarma, A. A. (2021b). POTENTIAL IN VITRO INHIBITORY EFFECTS OF MORINGA OLEIFERA LEAF EXTRACTS ON EXTENDED-SPECTRUM β-LACTAMASE-PRODUCING BACTERIA. Journal of Fundamental and Applied Sciences, 13(1), 137-150.
- Garba, L., Yushaâ, M., Abdullahi, M. M., Abubakar, M. U., Inuwa, A. B., Isa, S., & Adamu, M. T. (2018).
 Effectiveness of Double Discs Synergy Test in the Confirmation of Extended Spectrum β-lactamase (ESβL) Production. *Journal of Biochemistry, Microbiology and Biotechnology*, 6(2), 15-18.
- Gill, A. A., Singh, S., Thapliyal, N., & Karpoormath, R. (2019). Nanomaterial-based optical and electrochemical techniques for detection of methicillin-resistant Staphylococcus aureus: a review. *Microchimica Acta*, *186*, 1-19.
- Hapsari, A. P., Wahyuni, C. U., & Mudjianto, D. (2018). Knowledge of surveillance officers on identification of healthcare-associated infections in surabaya. *Jurnal Berkala Epidemiologi*, 6(2), 130.
- Heydarpour, F., Rahmani, Y., Heydarpour, B., & Asadmobini, A. (2017). Nosocomial infections and antibiotic resistance pattern in open-heart surgery patients at Imam Ali Hospital in Kermanshah, Iran. *GMS hygiene and infection control*, *12*.
- Hu, Z., Wang, Z., Liu, D., Chen, P., Wang, H., Chen, Y., & Shi, Y. (2011). Clinical and molecular microbiological characteristics of carbapenem-resistant *Acinetobacter baumannii* strains in an NICU. *Pediatrics International*, 53(6), 867-872.
- Ibrahim, M. E. (2019). Prevalence of *Acinetobacter baumannii* in Saudi Arabia: risk factors, antimicrobial resistance patterns and mechanisms of carbapenem resistance. *Annals of clinical microbiology and antimicrobials*, 18(1), 1.
- Jain, M., Sharma, A., Sen, M. K., Rani, V., Gaind, R., & Suri, J. C. (2019). Phenotypic and molecular characterization of *Acinetobacter baumannii* isolates causing lower respiratory infections among ICU patients. *Microbial pathogenesis*, 128, 75-81.
- Khaledi, M., Shahini Shams Abadi, M., Validi, M., Zamanzad, B., Vafapour, R., & Gholipour, A. (2019).
 Phenotypic and genotypic detection of metallo-β-lactamases in *A. baumanii* isolates obtained from clinical samples in Shahrekord, southwest Iran. *BMC Research Notes*, *12*(1), 1-6.

- Mahmood, N. H., & Al-Brefkani, A. (2022). Molecular Characterisation of Carbapenemase-Producing Acinetobacter baumannii isolates from Hospitalised Patients in Iraq. Journal of Life and Bio Sciences Research, 3(02), 27-32.
- Mayasari, E., & Siregar, C. (2015). Prevalence of *Acinetobacter baumannii* isolated from clinical specimens in adam malik hospital. *Majalah Kedokteran Andalas*, *37*(1), 1-7.
- Mirnejad, R., Moradli, G., Mirkalantari, S., & Golmohammadi, R. (2018). Molecular genotyping of *Acinetobacter baumannii* species isolated from patients in Tehran, Iran, by repetitive element PCR fingerprinting. *Iranian Journal of Pathology*, *13*(2), 144.
- Nemec A, Radolfova-Krizova L, Maixnerova M, Vrestiakova E, Jezek P, Sedo O (2016). Taxonomy of haemolytic and/or proteolytic strains of the genus Acinetobacter with the proposals of Acinetobacter courvalinii sp. nov. (Genomic species 14 sensu Bouvet & Jeanjean), Acinetobacter dispersus sp. nov.(genomic species 17), Acinetobacter modestus sp. nov., Acinetobacter proteolyticus sp. nov. and Acinetobacter vivianii sp. nov. International Journal of Systematic and Evolutionary Microbiology, 66(4):1673-1685
- Nimer N. A. (2022). Nosocomial Infection and Antibiotic-Resistant Threat in the Middle East. Infection and Drug Resistance, 15: 631-639
- Odewale, G., Adefioye, O. J., Ojo, J., Adewumi, F. A., & Olowe, O. A. (2016). Multidrug resistance of *Acinetobacter baumannii* in Ladoke Akintola university teaching hospital, Osogbo, Nigeria. *European Journal of Microbiology and Immunology*, 6(3), 238-243.
- Parajuli, N. P., Acharya, S. P., Mishra, S. K., Parajuli, K., Rijal, B. P., & Pokhrel, B. M. (2017). High burden of antimicrobial resistance among gram negative bacteria causing healthcare associated infections in a critical care unit of Nepal. *Antimicrobial Resistance & Infection Control*, 6(1), 1-9.
- Poorzargar, P., Javadpour, S., & Karmostaji, A. (2017). Distribution and antibiogram pattern of Acinetobacter infections in Shahid Mohammadi hospital, Bandar Abbas, Iran. Bimonthly Journal of Hormozgan University of Medical Sciences, 20(6), 396-403.
- Rizk, S. S., Elwakil, W. H., & Attia, A. S. (2021). Antibiotic-Resistant Acinetobacter baumannii in Low-Income Countries (2000–2020): Twenty-One Years and Still below the Radar, Is It Not There or Can They Not Afford to Look for It?. Antibiotics, 10(7), 764.
- Saba N., Balwan W.K. (2023). Study of frequency, epidemiology of nosocomial infections in healthcare centres. *Saudi Journal of Pathology and Microbiology*, 8(7):160-6.
- Shareef, S. M., & Risan, M. H. (2021). Molecular Identification of epsA and ompA genes for Acintobacter baumannii Isolated from Burn Wounds. Indian Journal of Forensic Medicine & Toxicology, 15(2), 841-848.
- Shi, X., Wang, H., Wang, X., Jing, H., Duan, R., Qin, S., & Wang, J. (2021). Molecular characterization and antibiotic resistance of *Acinetobacter baumannii* in cerebrospinal fluid and blood. *Plos one*, 16(2), e0247418.
- Tarigan, G. E., Nawan, N., & Toemon, A. I. (2023). Identification and Resistance Testing of Bacteria Causing Nosocomial Infections in Surgery Inpatient Rooms. *Disease Prevention and Public Health Journal*, 17(1), 100-108.
- Vaiyapuri, M., Joseph, T. C., Rao, B. M., Lalitha, K. V., & Prasad, M. M. (2019). Methicillin-resistant *Staphylococcus aureus* in seafood: prevalence, laboratory detection, clonal nature, and control in seafood chain. *Journal of food science*, 84(12), 3341-3351.

Yang W, Yao H, Xi C, Ye X, Chen Q, Zhang J, Yu J, Hu J. (2023). Prevalence and Clinical Characteristics of Bacterial Pneumonia in Neurosurgical Emergency Center Patients: A Retrospective Study Spanning 13 Years at a Tertiary Center. *Microorganisms*, 11(8):1992.