ORIGINAL ARTICLE

Antibacterial Susceptibility Patterns of Nonfermenting Gram-Negative Bacilli among Patients in a Tertiary Care Hospital, Jaipur

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Abstract. Introduction: *In recent decades, infections caused by Pseudomonas aeruginosa and Acinetobacter baumannii have also occurred outside the intensive care unit (ICU), affecting patients with comorbidities in the community. Most of the nonfermenting Gram-negative bacilli (NFGNB) cause nosocomial bloodstream infections, particularly in debilitated and immunocompromised hosts. Our study aimed to find out the antibacterial susceptibility patterns of NFGNB isolates in the clinical samples.* ***Materials and methods:*** *The study included all NFGNB isolates from various clinical samples that the clinical microbiology laboratory received from inpatients and outpatients at Mahatma Gandhi Medical College and Hospital in Jaipur, Rajasthan, India. Routine microscopy of samples was done. We performed Gram staining on all samples, with the exception of urine. We inoculated all clinical samples on blood agar and MacConkey agar, then incubated them at 37°C for 18-24 hours. Colony characteristics were observed. All organisms that generated pale or colorless colonies on MacConkey agar and exhibited Gram-negative bacteria upon Gram staining of the colony were classified as non-fermenting Gram-negative bacteria (NFGNB) and subsequently identified using the VITEK-2 compact system.* ***Results.*** *We identified 879 NFGNB isolates from a total of 10,707 clinical samples. Of these, 415 (47.21%) were Pseudomonas aeruginosa and 378 (43.2%) were Acinetobacter baumannii. The majority of isolates were from males in the age group of 61-70 years (13.76%), followed by the age group 41-50 years (12.85%). Pseudomonas aeruginosa was most commonly isolated from pus swab (13.42%), and Acinetobacter baumannii was isolated most commonly from endotracheal secretions (21.4%).* ***Conclusions.*** *Increasing antibiotic resistance will lead to challenges in managing all NFGNB unless appropriate measures are implemented and novel medications are developed. In order to prevent the spreading of resistant Pseudomonas aeruginosa and Acinetobacter strains, infection control measures should be taken, clinicians and laboratory workers should cooperate during antibiotic use, and hospital hygienic rules should be observed.*

Keywords*: nonfermenting Gram-negative bacilli; antimicrobial susceptibility; Pseudomonas aeruginosa; Acinetobacter baumannii*

1. INTRODUCTION

Nonfermenting Gram-negative bacilli (NFGNB) are a heterogeneous group of aerobic, non-sporing bacteria that do not metabolize glucose as an energy source or do it oxidatively. They represent approximately one-fifth of all Gram-negative bacilli (GNB) [1] and are found as saprophytes in nature, inhabiting soil and water, and also as commensals in people and animals. Despite their frequent isolation as incidental organisms, the advent of antibiotic resistance and patients with reduced immune responses have led to their increased frequency as pathogens. Non-fermenters (NFs)

are emerging with increasing frequency as agents of opportunistic and often serious infections as well as nosocomial infections [2]. NFGNB are most commonly isolated from patients with serious underlying disease who have abusive use of wide-spectrum antimicrobial agents, prolonged surgical procedures, prolonged hospital stays, inadequate mechanical instrumentation or tracheotomy, genitourinary instrumentation, in burn patients, and low birth weight babies. These infections can also be identified in extreme age groups like neonates, children, and geriatric age [3]. They are frequently isolated from patients with diseases like septicaemia, meningitis, pneumonia, urinary tract infections, and surgical wound infections [4]. NFGNB can cause a wide variety of infections and account for approximately 15% of all Gram-negative bacilli cultured from clinical samples [5].

The infections caused by NFGNB represent an emerging problem in nosocomial settings, especially in an immunocompromised host [6]. *Pseudomonas aeruginosa* is a prominent opportunistic pathogen in immunocompromised hosts due to disease or therapy, followed by *Acinetobacter baumannii, Pseudomonas fluorescens, Pseudomonas stutzeri, Stenotrophomonas maltophilia, Pseudomonas putida,* and *Burkholderia cepacia*. Colonization rates rise in hospitalized patients, especially those with prolonged stays or those who have undergone broad-spectrum antibiotic medication or chemotherapy. Most NFGNB are responsible for nosocomial bloodstream infections, particularly in debilitated and immunocompromised individuals, and are typically multidrug-resistant. Data from the Surveillance and Control of Pathogens of Epidemiological Importance (SCOPE) study revealed that approximately one fourth of Gram-negative bacteremia were attributed to NFGNB [8].

The infection caused by these agents is challenging to treat with antimicrobials due to the rapid selection of high-level multidrug resistance (MDR) to a variety of antibiotics, including β-lactams, aminoglycosides, and fluoroquinolones, which poses a problem for both treatment and infection control [9]. *Pseudomonas aeruginosa* has greater potential to become resistant to most antibiotics, as is clear from the fact that its genome contains the largest resistance island with more than 50 resistance genes. The mechanism of antibiotic resistance involves the synthesis of antibiotic-degrading or antibiotic-inactivating enzymes, outer membrane proteins that expel drugs, and mutations that alter antibiotic targets [10].

The production of antibiotic-degrading enzymes, including extended-spectrum beta-lactamases, AmpC cephalosporinases, carbapenemases, and aminoglycoside-modifying enzymes, has been documented in NFGNB [11]. NFGNB are resistant to many antibiotics and are known to produce extended-spectrum beta-lactamases and metallo-beta-lactamases [12]. Resistance compromises treatment, prolongs hospital stay, and increases mortality rate and healthcare costs [13].

The aim of our study was to assess the antibacterial susceptibility patterns of NFGNB isolates in the clinical samples.

2. MATERIALS AND METHODS

We conducted a prospective study at Mahatma Gandhi Medical College and Hospital in patients presenting signs and symptoms of non-fermenter infections from June 2019 to May 2020. The clinical microbiology laboratory at Mahatma Gandhi Hospital (MGH), Sitapura, Jaipur, Rajasthan, India, received all NFGNB isolates from various clinical samples from inpatients and outpatients.

Collection & Transportation of Specimen

Various sample types, such as urine, blood, pus, discharge from skin and soft tissue, sputum, endotracheal secretions, various fluids (including cerebrospinal fluid – CSF, ascitic fluid, pleural fluid, etc.), and various swabs (including ear, throat, vaginal, and wound swabs) were obtained from various clinical specimens received in the clinical microbiology laboratory. We collected samples with universal precautions, using prescribed sterile techniques and transported them to the laboratory as soon as possible, ensuring optimum transportation conditions. Detailed relevant history was taken as age, sex, the

history of any indwelling medical devices used, and duration of wards and ICU stay. We collected all the samples from a variety of inpatients and outpatients.

Transport and storage of specimen

Following sample collection, we properly labelled the container with the patient's name, ID number, and other details. We promptly transferred the specimens to the laboratory, typically within an hour of collection, and expedited their processing. If the processing took longer than expected, we stored them at 4^0C.

Processing of Specimen

Routine microscopy of specimens was conducted. Gram staining was performed on all samples except for urine. Wet microscopy of urine samples was used to examine bacteria and pus cells. All clinical samples were inoculated onto Blood agar and MacConkey agar and incubated at 37°C for 18-24 hours. Colony characteristics were examined. All organisms that produced pale or colorless colonies on MacConkey agar and exhibited Gram-negative bacilli on Gram staining of the colonies were classified as NFGNB and then identified using the VITEK-2 compact system [14]. All NFGNB isolates underwent antimicrobial susceptibility testing using the Kirby-Bauer Method [15].

3. RESULTS

The study was carried out at Mahatma Gandhi Medical College and Hospital in the Department of Microbiology. A total of 10707 samples were processed for bacterial culture. We identified 879 NFGNB isolates from a total of 10,707 clinical samples in our study. Out of 879 NFGNB isolates, 415 (47.21%) were identified as *Pseudomonas aeruginosa*, followed by *Acinetobacter baumannii* at 378 specimens (43.00%), *Burkholderia cepacia* at 29 specimens (3.30%), *Burkholderia pseudomallei* at 17 specimens (1.93%), *Stenotrophomonas maltophilia* at 16 specimens (1.82%), *Acinetobacter lwoffii* at 7 specimens (0.80%), *Pseudomonas putida* at 6 specimens (0.68%), *Acinetobacter coffee* at 2 specimens (0.23%), *Acinetobacter fwinii* at 2 specimens (0.23%), *Pseudomonas fluorescens* at 2 specimens (0.23%), *Pseudomonas stutzeri* at 2 specimens (0.23%), *Acinetobacter junii* at one specimen (0.11%), *Acinetobacter haemolyticus* at one specimen (0.11%), and *Pseudomonas luteola* at one specimen (0.11%), as detailed in Table 1.

Table 1. Total number of isolates obtained from NFGNB.

NFGNB spp.	Total number	Percentage
Pseudomonas aeruginosa	415	47.21%
Acinetobacter baumannii	378	43.00%
Burkholderia cepacia	29	3.30%
Burkholderia pseudomallei	17	1.93%
Stenotrophomonas maltophilia	16	1.82%
Acinetobacter lwoffii	7	0.80%
Pseudomonas putida	6	0.68%
Acinetobacter coffee	2	0.23%
Acinetobacter fwinii	2	0.23%
Pseudomonas fluorescens	2	0.23%
Pseudomonas stutzeri	2	0.23%
Acinetobacter junii	1	0.11%
Acinetobacter haemolyticus	1	0.11%
Pseudomonas luteola	1	0.11%
TOTAL	**879**	**100%**

Among the 879 isolates of NFGNB, the predominant specimens were from ET (30.60%), followed by pus swab (19.11%), blood (16.15%), and urine (10.01%), as illustrated in Table 2.

Table 2. Various clinical specimens collected in the study (ET - endotracheal secretions, CSF cerebrospinal fluid, BAL – bronchoalveolar lavage).

Sl. No.	Sample	No. of cases	Percentage
1	ET	269	**30.60%**
2	PUS SWAB	168	**19.11%**
3	BLOOD	142	**16.15%**
4	URINE	88	**10.01%**
5	SPUTUM	87	9.90%
6	CSF	44	5.01%
7	PUS	19	2.16%
8	DRAIN	16	1.82%
9	PLEURAL FLUID	11	1.25%
10	BAL	11	1.25%
11	TIP	6	0.68%
12	TISSUE	5	0.57%
13	ASCITIC FLUID	4	0.46%
14	OTHER BODY FLUID	3	0.34%
15	SYNOVIAL FLUID	2	0.23%
16	SEMEN	2	0.23%
17	BILE	2	0.23%
	TOTAL	**879**	**100%**

Among the total positive NFGNB cases, the majority were from inpatient department (IPD) with 773 cases (87.94%), followed by outpatient department (OPD) with 106 cases (12.06%). Among the 879 NFGNB isolates, the predominant demographic was males aged 61-70 years, including 121 isolates (13.76%), followed by those aged 41-50 years, with 113 isolates (12.85%), as illustrated in Table 3 and Figure 1.

Table 3. Distribution of positive NFGNB isolates with respect to outpatient department/inpatient department.

Distribution of OPD/IPD	Number of Isolates	Percentage
OPD	106	12.06%
IPD	773	87.94%
Total	879	100%

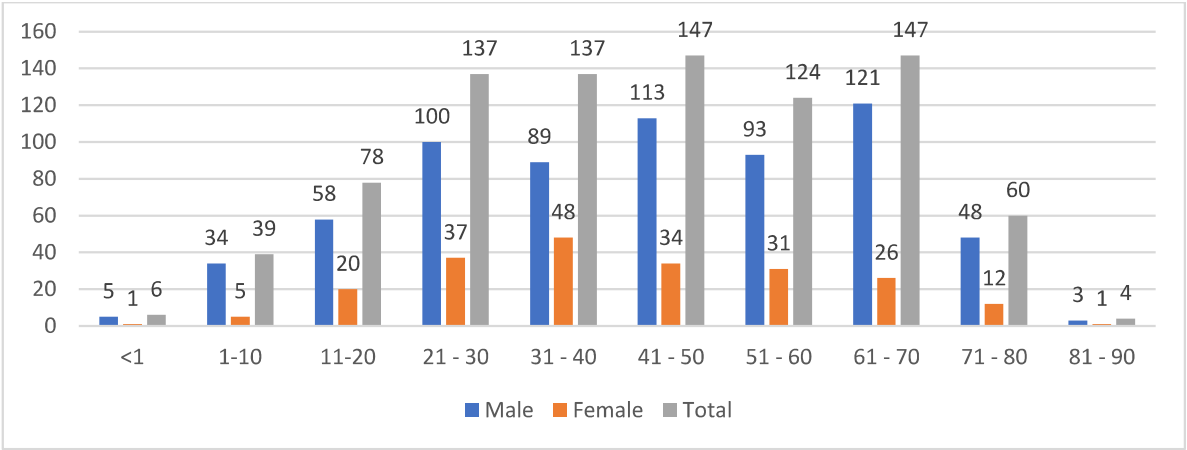

Figure 1. Gender-wise distribution of patients.

Table 4 depicts the most common isolates from various clinical samples. In the current investigation, *Pseudomonas aeruginosa* was predominantly isolated from pus swabs (13.42%), followed by ET (8.19%), sputum (7.17%), urine (6.94%), and blood (4.44%). *Acinetobacter baumannii* was predominantly isolated from ET (21.4%), followed by blood (7.85%), pus swabs (4.44%), CSF (3.53%), sputum (2.05%), and urine (1.82%). *Burkholderia cepacia* was predominantly isolated from blood (1.48%), followed by pus swab (0.68%), sputum (0.34%), ET samples (0.23%), and other bodily fluids (0.23%).

Table 4. Distribution of microorganism on the bases of specimens (ET - endotracheal secretions, CSF cerebrospinal fluid, BAL – bronchoalveolar lavage).

SAMPLE	P. AERUGINOSA		A. BAUMANNII		B. CEPACIA	
	N	%	N	%	N	%
ET (n=269)	72	8.19	188	21.39	2	0.23
PUS SWAB (n=168)	118	13.42	39	4.44	6	0.68
BLOOD (n=142)	39	4.44	69	7.85	13	1.48
URINE (n=88)	61	6.94	16	1.82	1	0.11
SPUTUM (n=87)	63	7.17	18	2.05	3	0.34
CSF (n=44)	12	1.37	31	3.53	0	0
PUS (n=19)	14	1.59	2	0.23	1	0.11
DRAIN (n=16)	12	1.37	3	0.34	0	0
PLEURAL FLUID (n=11)	5	0.57	4	0.46	0	0
BAL (n=11)	5	0.57	5	0.57	0	0
TIP (n=6)	5	0.57	0	0	1	0.11
TISSUE (n=5)	3	0.34	2	0.23	0	0
ASCITIC FLUID (n=4)	2	0.23	1	0.11	0	0

OTHER BODY FLUID (n=3)	0	0	0	0	2	0.23
SYNOVIAL FLUID (n=2)	0	0	0	0	0	0
SEMEN (n=2)	2	0.23	0	0	0	0
BILE (n=2)	2	0.23	0	0	0	0
TOTAL	415	47.21	378	43	29	3.3

Antimicrobial susceptibility testing was conducted on all 879 NFGNB, revealing a sensitivity of 37.54% to Cefoperazone/Sulbactam, followed by Meropenem (35.26%), Ceftazidime (34.47%), Gentamicin (33.33%), and Netilmicin (33.21%). Resistance rates were notably high, with 90.78% resistance to Ticarcillin/Clavulanic Acid, followed by Ciprofloxacin (73.49%), Levofloxacin (72.81%), Piperacillin/Tazobactam (70.19%), and Amikacin (69.16%), as illustrated in Table 5.

Table 5. Antimicrobial susceptibility pattern of NFGNB.

S. No.	**Antibiotics**	**Sensitive**		**Resistance**	
		No. of cases	**%**	**No. of cases**	**%**
1	Ticarcillin/Clavulanic Acid	81	9.21%	798	90.78%
2	Piperacillin/Tazobactam	262	29.80%	617	70.19%
3	Ceftazidime	303	34.47%	576	65.52%
4	Cefoperazone/Sulbactam	330	37.54%	549	62.45%
5	Cefepime	288	32.76%	591	67.23%
6	Imipenem	285	32.42%	594	67.57%
7	Meropenem	310	35.26%	569	64.73%
8	Amikacin	271	30.83%	608	69.16%
9	Gentamycin	293	33.33%	586	66.66%
10	Netilmicin	292	33.21%	587	66.78%
11	Ciprofloxacin	233	26.50%	646	73.49%
12	Levofloxacin	239	27.18%	640	72.81%

A total of 879 NFGNB isolates were collected, among which 415 isolates of *P. aeruginosa* exhibited high sensitivity to cefepime (258, 62.16%), followed by amikacin (257, 61.92%), imipenem (254, 61.20%), and ceftazidime (250, 60.24%). Conversely, there was significant resistance to ticarcillin/clavulanic acid (349, 84.09%), followed by levofloxacin (226, 54.45%) and ciprofloxacin (208, 50.12%).

A total of 378 *Acinetobacter baumannii* isolates exhibited significant sensitivity to cefoperazone/sulbactam, with 68 isolates (17.98%) demonstrating sensitivity, followed by netilmicin with 51 isolates (13.49%), and gentamicin with 35 isolates (9.25%).

Conversely, there was a pronounced resistance to amikacin, with 375 isolates (99.20%), followed by ticarcillin/clavulanic acid with 366 isolates (96.82%), and ciprofloxacin with 363 isolates (96.03%).
Burkholderia cepacia isolates exhibited significant sensitivity to meropenem (65.51%, n = 29), followed by ceftazidime (48.27%) and levofloxacin (37.93%). Conversely, the isolates demonstrated resistance to ticarcillin/clavulanic acid, piperacillin/tazobactam, cefepime, imipenem, amikacin, and gentamicin.

Table 6. Antimicrobial susceptibility pattern of isolated NFGNB species.

ANTIBIOTIC		*P. aeruginosa* (n=415)		A. BAUMANNII (n=378)		B. CEPACIA (n=29)	
		N	%	N	%	N	%
Ticarcillin/Clavulanic Acid	R	349	84.09%	366	96.82%	29	100.00%
	S	66	15.90%	12	3.17%	0	0.00%
Piperacillin/Tazobactam	R	185	44.57%	357	94.44%	29	100.00%
	S	230	55.42%	21	5.55%	0	0.00%
Ceftazidime	R	165	39.75%	363	96.03%	15	51.72%
	S	250	60.24%	15	3.96%	14	48.27%
Cefoperazone/Sulbactam	R	169	40.72%	310	82.01%	28	96.55%
	S	246	59.27%	68	17.98%	1	3.44%
Cefepime	R	157	37.83%	358	94.70%	29	100.00%
	S	258	62.16%	20	5.29%	0	0.00%
Imipenem	R	161	38.79%	358	94.70%	29	100.00%
	S	254	61.20%	20	5.29%	0	0.00%
Meropenem	R	167	40.24%	360	95.23%	10	34.48%
	S	248	59.75%	18	4.76%	19	65.51%
Amikacin	R	158	38.07%	375	99.20%	29	100.00%
	S	257	61.92%	3	0.79%	0	0.00%
Gentamycin	R	169	40.72%	343	90.74%	29	100.00%
	S	246	59.27%	35	9.25%	0	0.00%
Netilmicin	R	184	44.33%	327	86.50%	28	96.55%
	S	231	55.66%	51	13.49%	1	3.44%
Ciprofloxacin	R	208	50.12%	363	96.03%	28	96.55%
	S	207	49.87%	15	3.96%	1	3.44%
Levofloxacin	R	226	54.45%	359	94.97%	18	62.06%
	S	189	45.54%	19	5.02%	11	37.93%

4. DISCUSSION

Pseudomonas aeruginosa and *Acinetobacter baumannii* are the most often reported microorganisms globally and have emerged as some of the most resistant healthcare-associated diseases to manage and cure. Patients admitted to the burn unit, intensive care unit, and those with central intravenous catheters and respiratory devices are the primary targets of this microorganism.
In the present study, a total of 10,707 specimens were received in the microbiology laboratory for culture. Among these samples, 879 were identified as positive isolates of NFGNB. Among them *Pseudomonas aeruginosa* was the most commonly isolated (47.21%), followed by *Acinetobacter baumannii*

(43.00%), *Burkholderia cepacia* (3.30%), *Burkholderia pseudomallei* (1.93%), *Stenotrophomonas maltophilia* (1.82%), *Acinetobacter lwoffii* (0.80%), *Pseudomonas putida* (0.68%), *Acinetobacter coffee* (0.23%), *Acinetobacter fwinii* (0.23%), *Pseudomonas fluorescens* (0.23%), *Pseudomonas stutzeri* (0.23%), *Acinetobacter junii* (0.11%), *Acinetobacter haemolyticus* (0.11%) and *Pseudomonas luteola* (0.11%). Our findings align with those reported by Simgamsetty et al., who isolated 260 NFGNB, comprising 151 (58%) *Pseudomonas aeruginosa*, 94 (36.1%) *Acinetobacter baumannii*, 5 (1.92%) *Burkholderia cepacia*, 3 (1.15%) *Acinetobacter lwoffii*, 3 (1.15%) *Pseudomonas stutzeri*, 1 (0.38%) *Achromonas xylosoxidans*, and one (0.38%) *Sphingomonas paucimobilis* [16]. Maniyan et al., Prashant et al., and Mahajan reported similar results [17-19].

In our study, of the total 879 isolates of NFGNB, the majority of specimens were obtained from ET (30.60%), followed by pus swabs, blood, and urine. Our findings align with those published by Wadhwa et al., indicating that the majority of clinical specimens were derived from ET (67.44%), followed by sputum (9.30%), and BAL (8.72%), which is consistent with additional investigations conducted by H Patel et al. [20,21].

In the present study, of the total 879 (100%) positive NFGNB isolates, 773 (87.94%) were obtained from IPD and 106 (12.06%) from OPD. These results align with those of Nazir et al., who discovered that among 120 strains of NFGNB, 91 (75.8%) were isolated from IPD and 29 (24.16%) from OPD, corroborating the findings of Rashid et al. [22,23].

In the current investigation, the majority of isolates were from males in the age category of 61-70 years, with 121 patients (13.76%), followed by the age group of 41-50, with 113 patients (12.85%). The current study aligned with Mahajan et al., indicating that the majority of patients (35.13%) were aged between 40 and 60 years. Among the 342 NFGNB isolates, 59.79% were obtained from males and 40.20% from females. In the research by Nazir et al., males were more frequently afflicted than females in the under-10 age group (34.1%), followed by the over-60 age group (32.5%) [19,22].

We identified *Pseudomonas aeruginosa* predominantly in pus swabs, followed by ET, sputum, urine, and blood. The study conducted by Rashid et al. indicated that 10.5% of *Pseudomonas aeruginosa* isolates were obtained from wound swab samples. Kumar et al. conducted a comparable study, which revealed that pus was the primary source of *Pseudomonas aeruginosa* isolation (58%), with urine, stool, and other sources following suit. A comparable investigation was conducted by Mahajan and Madkey [19,23-25].

In the current investigation, *Acinetobacter baumannii* was predominantly isolated from ET (21.39%), followed by pus swabs, cerebrospinal fluid, sputum, and urine. The results were analogous to those reported by Madkey et al., who identified 58.82% *Acinetobacter baumannii* isolates from endotracheal samples, followed by pus, urine, blood, and sputum. Additional research was conducted by Mahajan et al. and Maniyan et al. [17,19,25].

In our investigation, *Burkholderia cepacia* was most frequently isolated from blood followed by pus swabs, sputum, and ET samples. These studies were consistent with those of other researchers who reported 5.00% of *Burkholderia cepacia* isolates from blood, followed by sputum [17,25,26].

In the current study, of the 879 NFGNB, 37.54% shown sensitivity to cefoperazone/sulbactam, followed by meropenem (35.26%), ceftazidime (34.47%), gentamicin (33.33%), and netilmicin (33.21%). These results aligns with the findings of Bhargava et al. and Sarkar et al., which indicated that NFGNB exhibit the most frequent sensitivity to cefoperazone/sulbactam [27,28].

In the current investigation, 258 isolates of *Pseudomonas aeruginosa* showed high sensitivity to cefepime, specifically 62.16%. This was followed by amikacin (61.92%), imipenem (61.20%), and ceftazidime (60.24%). The findings correspond with the study by Malini et al., which reported

sensitivity to cefepime in 38.33% of cases, to amikacin in 71.67% of cases, to imipenem in 86.67% of cases, and to ceftazidime in 33.39% of cases. These findings align with previous studies indicating that *Pseudomonas aeruginosa* has great sensitivity to cefepime, followed by amikacin and imipenem [29].

Out of 378 *Acinetobacter baumannii* isolates, 68 exhibited high sensitivity to cefoperazone/sulbactam (17.98%), 51 to netilmicin (13.49%), and 35 to gentamicin (9.25%). A comparable study conducted by Maniyan et al. indicated that *Acinetobacter baumannii* showed sensitivity to gentamicin in 18 cases (50%) [17].

In our study, of the 29 *Burkholderia cepacia* isolates, 19 (65.51%) exhibited strong sensitivity to meropenem, followed by 14 (48.27%) to ceftazidime and 11 (37.93%) to levofloxacin. The results closely resembled those of the studies conducted by Sharma et al., Simgamsetty et al., and Kalidas et al. [16,26,30]. To avoid the transmission of infection, control measures must be implemented, and physicians should adhere to hospital hygiene protocols.

5. CONCLUSIONS

NFGNB are important bacteria causing both hospital- and community-acquired infections. NFGNB are now emerging as important pathogens causing a wide range of nosocomial infections. *Pseudomonas aeruginosa* and *Acinetobacter baumannii* were the most common NFGNB isolated in our study. The accurate identification of NFGNB to the species level, together with the monitoring of their antibiotic resistance patterns, is essential for effective management of the infections they cause. Increased isolation and elevated antibiotic resistance are concerning indicators for healthcare workers. These pathogens can survive in hospital environment that's why proper housekeeping, equipment decontamination and strict guidelines for sterilization need to be implemented. Further studies will definitely help in better understanding of changes in its antimicrobial resistance pattern.

Author Contributions:
S.K, V.P.M and R.S. conceived the original draft preparation. S.K, V.P.M. and R.S. were responsible for the data acquisition, collection and assembly of the articles. S.K, V.P.M. and R.S. were responsible for the conception and design. S.K, V.P.M. and R.S. were responsible with the supervision of the manuscript

Compliance with Ethics Requirements:
"The authors declare no conflict of interest regarding this article".
"The authors declare that all the procedures and experiments of this study respect the ethical standards in the Helsinki Declaration of 1975, as revised in 2008(5), as well as the national law. Informed consent was obtained from all the patients included in the study"
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References

[1] Koneman EW, Alen SD, Janda WM, Schreckenbeiger PC, Winn WC. The non fermenting Gram negative bacilli. In: Colour Atlas and text book of Diagnostic Microbiology.5th edition, Philadelphia: J.B. Lippincott, 1997;253-309.

[2] Mishra E, Bhujwala RA, Shriniwas. Nonfermenters in human infection. *Indian J Med Res*. 1986;83:561-566.

[3] Gardener P, Griffin WB, Swartz MN, Kunz LJ. Non fermentative Gram-negative bacilli of nosocomial interest. *Amer J Med.* 1970;48(6):735-749.

[4] Kiska DL, Gilligan PH. Pseudomonas. In: Murray PR, Baron Jorgensen JH, Pfaller MA, Yolken RH, editors. Manual of Clinical Microbiology. 8th edition Washington DC: ASM Press. 2003;2:719-728.

[5] Siou CS, Mario V, Lenie D, Yu Fang Wei, Ya Lei Chen, Chang TC. Identification of non-fermenting Gram negative bacteria of clinical importance by an oligonucleotide array. *J Med Microbiol*. 2009;58(5):596-605.

[6] Koneman E, Allen S, Janda W, Schreckenberger P, Winn W. Colour Atlas and textbook of Diagnostic Microbiology. 6th Edition. Philadelphia: Lippincott -Raven Publishers. 2006:305-91.

[7] Govan JRW. *Pseudomonas*, *Stenotrophomonas, Burkholderia.* In: Collee JG, Fraser AG, Marimion, Simmons A, editors. Practical Medical Microbiology.14th edition, India: Churchchill Livingstone 2006; 448-461.

[8] Wisplinghoff H, Bischoff T, Tallent SM, Seifert H, Wenzel RP, Edmond MB. Nosocomial bloodstream infections in US hospitals: analysis of 24,179 cases from a prospective nationwide surveillance study. *Clin Infect Dis.* 2004;39(3):309-17.

[9] Taneja N, Maharwal S, Sharma M. Imipenem resistant in non-fermenters causing nosocomial urinary tract infection. *Ind J Med Sci.* 2003;57(7):294-299.

[10] Hancock REW. Resistance mechanisms in *Pseudomonas aeruginosa* and other nonfermentive Gram negative bacteria. *Clin Infect Dis*. 1998;27(1):93-99.

[11] Bokaeian M, Amini BN, Sahi Z, Saeidi S. Antibacterial activities of Myrtus communis L extract against multi-drug resistant *Klebsiella pneumoniae* and *Pseudomonas aeruginosa*. *Inter J Biosciences.* 2014;4(9):254-259.

[12] Yano H, Kuga A, Okamoto R, Kitasato H, Kobayashi T, Inone M. Plasmid encoded metallo- beta-lactamase (IMP-6) conferring resistance to carbapenems, especially meropenem. *Antimicrob Agents Chemother*. 2001;45(5):1343-8.

[13] Kurokawa H, Yagi T, Shibata N, Shibayana K, Arakawa Y. Worldwide proliferation of carbapenem resistant Gram-negative bacteria. *Lancet* 1999;354(9182):955.

[14] Joyanes P, del Carmen Conejo M, Martinez-Martinez L, Perea E J. Evaluation of the VITEK 2 system for the identification and susceptibility testing of three species of nonfermenting gram negative rods frequently isolated from clinical samples. *J Clin Microbiol* 2001;39(9): 3247-3253.

[15] James J, Biemer, M.D. St. Antimicrobial susceptibility testing by the Kirby-Bauer Disc Diffusion method annals of Clinical *Laboratory Science*. 1973;3(2).135-140.

[16] Simgamsetty S, Yarlagadda P, Myneni RB, Penmetcha U. Identification and antimicrobial susceptibility patterns of nonfermentive Gram negative bacilli in a tertiary care hospital. *International Journal of Health Sciences and Research* 2016;6(8):105-111.

[17] Maniyan G, Vedachalam D, Chinnusamy N. Characterization and antimicrobial susceptibility pattern of non-fermenting Gram negative bacilli from various clinical samples in a tertiary care hospital. *Indian J Microbiol Res*. 2016;3(4):387-391.

[18] Prashant K, Parandekar, Peerapur BV. Non-fermenters in human infections with special reference to *Acinetobacter Species* in a tertiary care hospital. *Journal of Krishna Institute of Medical Sciences University* 2012;1(1):84-88.

[19] Mahajan R, Sarika N, Mahajan B. Isolation and identification of non fermenting Gram negative bacilli in a tertiary care hospital. *Sch J App Med Sci.* 2016; 4(3D):872-876.

[20] Wadhwa R, Sharma Y, Pandeyupadhyay R, Bala K. Nosocomial infection by nonfermenting Gram negative bacilli in tertiary care hospital: screening and cure. *Int J Pharm Pharm Sci*. 2016;8(3):274-277.

[21] Patel PH, Pethani JD, Rathod SD, Chauhan B, Shah PD. Prevalence of non-fermenting Gram negative bacilli infection in tertiary care Hospital in Ahmedabad, Gujarat. *Indian Journal of Basic Applied Medical Research*. 2013;6(2):608 -613.

[22] Nazir A, Peerzada BY, Sana I. Spectrum of non-fermenting Gram negative bacilli isolated from patients with blood stream infections in a tertiary care hospital in North India. *Int J Res Med Sci.* 2019;7(5):1762-1766.

[23] Rashid A, Chowdhury A, Rahman S, Begum S, Muazzam N. Infections by *Pseudomonas aeruginosa* and antibiotic resistance pattern of the isolates from Dhaka Medical College Hospital'. Bangladesh. *J Med Microbiol* 2007;1(2):48-51.

[24] Sarkar AK, Shaker M, Fasle RD. Isolation identification and antibiotic susceptibility test of non-fermenters from different clinical specimens. *Journal of Advances in Applied Sciences and Technology* 2014;1(1):48-52.

[25] Madkey MV, Gajbhiye SR. Study of nonfermentive Gram negative bacilli. *International Journal of Basic and Applied Research* 2019;10(5):e5189.

[26] Kalidas RIT, Falguni N, Hirak JR, Maity PK. Prevalence and susceptibility profiles of non-fermentative gram- negative bacilli infection in a tertiary care hospital of eastern India. *Indian J Clin Practice* 2013;24(5):451-5.

[27] Bhargava D, Kar S, Saha M. Prevalence of non-fermentative Gram negative bacilli infection in tertiary care hospital in Birgunj, Nepal. *Int J Curr Microbiol App Sci.* 2015;4(7):301-307.

[28] Sarkar M, Jena J, Pattnaik D, Mallick B. Prevalence of nonfermentative Gram-negative bacilli and their antimicrobial susceptibility profiles in a tertiary care hospital of Eastern India. *Int J Adv Med.* 2018;5(2):366-370.

[29] Malini A, Deepa EK, Gokul BN, Prasad SR; Nonfermenting Gram-negative bacilli infections in a tertiary care hospital in Kolar, Karnataka. *Journal of Laboratory Physicians.* 2009;1(2):62-66.

[30] Sharma D, Vyas N, Sinha P, Mathur A. Non fermentative Gram negative bacilli as nosocomial pathogens: Identification and antibiotic sensitivity in clinical samples of indoor patients. *Nepal Journal of Medical Sciences.* 2014;3(2):101-105