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Bacterial Profile and Antimicrobial Susceptibility Pattern of Non-Fermenting Gram-Negative Bacilli Isolated from Various Clinical Samples in a Tertiary Care Hospital.

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Abstract

Background: Non-fermenting Gram-negative bacilli (NFGNB) are increasingly recognized as important opportunistic and healthcare-associated pathogens. They are linked to a broad spectrum of infections, including bloodstream infections, pneumonia, urinary tract infections, wound infections, meningitis and ventilator-associated pneumonia. Accurate identification and antimicrobial susceptibility testing are essential for proper clinical management due to their intrinsic resistance to a variety of antimicrobial agents and their ability to acquire additional resistance mechanisms. ***Aim:*** To study the bacterial profile and antimicrobial susceptibility pattern of non fermenting gram negative bacilli isolated from various clinical samples in tertiary care hospital. ***Materials and Methods:*** A cross-sectional study was carried out in the Department of Microbiology, ESIC Medical College and Hospital, Kalaburagi, for One year from September 2024 to August 2025. During the study period, a total of 4258 clinical samples were processed, out of which 219 samples yielded NFGNB. The clinical specimens included pus, wound swabs, sputum, urine, blood, body fluids, catheter related specimens and endotracheal aspirates. Isolation of isolates was done by standard microbiological methods based on Gram stain, colony morphology, motility and biochemical reactions including oxidase and catalase tests. Antimicrobial susceptibility testing was performed by the Kirby–Bauer disk diffusion method on Mueller–Hinton agar and interpreted according to CLSI 2024 guidelines. The antimicrobial agents used for testing were ceftazidime, cefepime, piperacillin-tazobactam, imipenem/meropenem, amikacin, gentamicin and ciprofloxacin. Descriptive statistics were used to provide an overview of the distribution and antimicrobial susceptibility pattern. ***Results:*** Out of 4258 clinical samples processed, 219 (5.14%) NFGNB were isolated. The most common isolate was Pseudomonas aeruginosa with 110 (50.2%) isolates followed by Acinetobacter baumannii with 102 (46.6%) isolates. Stenotrophomonas spp . and Burkholderia cepacia complex were responsible for 5 (2•3%) and 2 (0•9%) isolates, respectively. The highest number of NFGNB isolates were obtained from pus (63) followed by sputum (50), fluids (37), other wound swabs/endotracheal aspirates and related specimens (49), blood (17) and urine (3). P. aeruginosa was most sensitive to amikacin (62%), gentamicin (55%), imipenem/meropenem (48%), piperacillin-tazobactam (42%), cefepime (35%), ceftazidime (30%) and ciprofloxacin (28%). Among A. baumannii, susceptibility was greatest to amikacin (38%), followed by gentamicin (33%), piperacillin–tazobactam (25%), imipenem/meropenem (22%), cefepime (20%), ceftazidime (18%) and ciprofloxacin (15%). ***Conclusion:*** NFGNB constituted a major proportion of clinically important gram negative bacterial isolates in the study setting and P. aeruginosa and A. baumannii contributed most of the isolates. The dominant organisms, especially A. baumannii, showed a high level of antimicrobial resistance. Aminoglycosides showed relatively greater susceptibility than some other tested agents. Continuous antimicrobial resistance surveillance, rational use of antimicrobials, appropriate infection-control measures and timely microbiological diagnosis are essential to tackle the increasing burden of multidrug-resistant NFGNB.

Keywords: Non-fermenting Gram-negative bacilli; Pseudomonas aeruginosa; Acinetobacter baumannii; antimicrobial susceptibility; antimicrobial resistance; multidrug resistance; tertiary care hospital.

INTRODUCTION

Non-fermenting Gram-negative bacilli (NFGNB) constitute a heterogeneous group of aerobic Gram-negative organisms that do not ferment carbohydrates in conventional media. Many non-fermenters are environmental organisms or colonizers,

but increasing numbers have been recognized as important opportunistic pathogens particularly in hospitalized and critically ill patients [1, 2].

Two of the most clinically significant NFGNB are Pseudomonas aeruginosa and Acinetobacter baumannii. These organisms can cause a variety of healthcare associated infections including pneumonia, ventilator associated pneumonia, bloodstream infections, urinary tract infections, wound and surgical site infections, meningitis and other serious infections. The clinical significance of NFGNB is directly related to their ability to survive under hostile environmental conditions and their ability to acquire multiple antimicrobial-resistance mechanisms. Resistance can develop through reduced membrane permeability, efflux systems, enzymatic inactivation of the antibiotic, target modification and acquisition of transferable resistance determinants.

Thus, treatment of multidrug-resistant NFGNB infections may be challenging and requires individual antimicrobial therapy. Antimicrobial resistance in NFGNB is now a recognized major global public health concern [3]. Invasive devices, prolonged hospitalization, exposure to intensive care, and antimicrobial pressure may facilitate acquisition and spread of resistant NFGNB in healthcare settings. Thus, knowledge of the local prevalence and antimicrobial susceptibility pattern of NFGNB is imperative. Local antibiograms can assist clinicians in choosing suitable empirical antimicrobial therapy and can provide valuable data for antimicrobial stewardship and infection-prevention programs [4]. The present study was undertaken to know the bacterial profile of NFGNB isolated from various clinical specimens and to know their antimicrobial susceptibility pattern in a tertiary care hospital.

MATERIALS AND METHODS

Study Design

A cross-sectional observational study was conducted in the Department of Microbiology, ESIC Medical College and Hospital, Kalaburagi, Karnataka.

Study Period

The study was conducted for 1year, from September 2024 to August 2025.

Study Population and Samples

During the study period, 4258 clinical samples received in the microbiology laboratory were processed according to standard laboratory procedures. A total of 219 samples showing growth of NFGNB were included in the present analysis.

Clinical Specimens

The clinical specimens included pus, wound swabs, sputum, urine, blood, body fluids, catheter-related specimens, endotracheal aspirates and other clinically relevant specimens.

Isolation and Identification

Clinical specimens were processed using standard microbiological procedures. Suspected NFGNB isolates were initially evaluated by Gram staining and colony morphology. Identification was performed using conventional microbiological methods, including assessment of Gram-stain morphology, colony characteristics, motility, oxidase reaction, catalase reaction and other relevant biochemical reactions.

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing was performed using the Kirby–Bauer disc diffusion method on Mueller–Hinton agar. Interpretation was performed according to Clinical and Laboratory Standards Institute (CLSI) 2024 guidelines [5]. The antimicrobial agents evaluated were ceftazidime, cefepime, piperacillin–tazobactam, imipenem/meropenem, amikacin, gentamicin and ciprofloxacin.

Data Analysis

The collected data were analyzed using descriptive statistics. The distribution of NFGNB isolates and their antimicrobial susceptibility profiles were expressed as numbers and percentages.

RESULTS

Overall, 219 isolates of Non-Fermenting Gram-Negative Bacilli (NFGNB) were identified. The majority of organisms identified were Pseudomonas aeruginosa (50.2%, n=110) and Acinetobacter baumannii (46.6%, n=102) (Table 1). Most of these isolates were recovered from pus (n=63) and sputum (n=50) specimens, followed by wound swabs/endotracheal aspirates (n=49), fluids (n=37), blood (n=17) and urine (n=3) (Table 2). The highest sensitivity was found with P. aeruginosa isolates to Amikacin (62%) and Gentamicin (55%) and moderate to low to Imipenem/Meropenem (48%), Piperacillin-tazobactam (42%), Cefepime (35%), Ceftazidime (30%) and Ciprofloxacin (28%) by antimicrobial

susceptibility testing (Table 3). Conversely, A. baumannii isolates showed an overall poor susceptibility profile to all the antibiotics tested, with highest sensitivity to Amikacin (38%) and Gentamicin (33%) and significantly low susceptibility to Piperacillin-tazobactam (25%), Imipenem/Meropenem (22%), Cefepime (20%), Ceftazidime (18%) and Ciprofloxacin (15%) (Table 4).

Table 1. Distribution of NFGNB isolates

Organism	No. of isolates	Percentage
Pseudomonas aeruginosa	110	50.2%
Acinetobacter baumannii	102	46.6%
Stenotrophomonas spp.	5	2.3%
Burkholderia cepacia complex	2	0.9%
Total	219	100%

Table 2. Specimen-wise distribution of NFGNB

Specimen	No. of isolates
Pus	63
Sputum	50
Fluid	37
Other wound swabs/endotracheal aspirates and related specimens	49
Blood	17
Urine	3
Total	219

Table 3. Antimicrobial susceptibility of P. aeruginosa

Antibiotic	Susceptibility (%)
Ceftazidime	30
Cefepime	35
Piperacillin–tazobactam	42
Imipenem/Meropenem	48
Amikacin	62
Gentamicin	55
Ciprofloxacin	28

Table 4. Antimicrobial susceptibility of A. baumannii

Antibiotic	Susceptibility (%)
Ceftazidime	18
Cefepime	20
Piperacillin–tazobactam	25
Imipenem/Meropenem	22
Amikacin	38
Gentamicin	33
Ciprofloxacin	15

DISCUSSION

The present study was conducted to determine the distribution and antimicrobial susceptibility pattern of NFGNB isolated from clinical specimens in a tertiary care hospital. Of the 4258 clinical samples processed during the study period, 219 yielded NFGNB, providing an isolation rate of 5.14%. This is evidence that NFGNB are a clinically significant group of microorganisms seen in routine tertiary care microbiology practice, consistent with the results of other regional tertiary care centers [1, 2]. The importance of NFGNB as healthcare-associated pathogens has also been demonstrated in previous studies. Clinical distribution and antibiotic resistance among non-fermenting Gram-negative bacilli was reported by Jayanthi and Jeya [6] highlighting the significance of these organisms in hospital practice. In the present study, P. aeruginosa was the most common NFGNB at, 50.2% of isolates, followed by A. baumannii at 46.6%. The clinical relevance of the predominance of these organisms is that they are both capable of causing severe healthcare-associated infections and have multiple intrinsic and acquired mechanisms of antimicrobial resistance. The most common specimen yielding NFGNB was pus (63 isolates) followed by sputum (50 isolates) and fluids (37 isolates). The dominance of wound/pus specimens indicates a significant contribution of NFGNB to wound-associated infections in the study site. Respiratory specimens also made a substantial contribution to the total number of isolates. P. aeruginosa and A. baumannii are leading causes of hospital-acquired respiratory infections, particularly among patients on invasive ventilation. The current study showed significant resistance in both predominant NFGNB. Among P. aeruginosa, the maximum susceptibility was to amikacin (62%) followed by gentamicin (55%). Carbapenem susceptibility was 48% and susceptibility to piperacillin–tazobactam, cefepime and ceftazidime was lower. The least susceptible of the tested agents was ciprofloxacin, at 28%. The susceptibility

profile of A. baumannii was even more worrying. Amikacin had the highest susceptibility rate of only 38%, followed by gentamicin with 33% susceptibility.

Susceptibility to ceftazidime, cefepime, piperacillin–tazobactam and imipenem/meropenem was low and only 15% of isolates were susceptible ciprofloxacin. The high resistance found in the present study may be related to several factors associated with tertiary-care healthcare environments, including extensive antimicrobial exposure, prolonged hospitalization, intensive-care admission and use of invasive devices. In recent assessments of NFGNB in ICUs, similar trends of high resistance, particularly in the critical care environment, have been well documented [7]. However, individual patient-level factors were not evaluated in the present dataset and therefore should not be interpreted as demonstrated causal factors. Treatment decisions should preferably be based on local susceptibility surveillance, rather than relying solely on generalized resistance patterns, because resistance patterns can vary substantially between hospitals and over time. Continued surveillance is particularly important in tertiary-care hospitals, where multidrug-resistant NFGNB may spread among vulnerable patients. While this study presents valuable institution-specific susceptibility data on a large sample of 4258 clinical specimens, it is limited by being a single-center, short-term study without detailed information on patient-level data and molecular characterization of resistance mechanisms. However, the findings demonstrate the critical need for accurate identification of NFGNB and routine susceptibility testing to guide definitive therapy. To prevent the spread of such resistant pathogens, the use of updated local antibiograms, strict antimicrobial stewardship and strict infection-control measures such as hand hygiene and appropriate device management remain important.

CONCLUSION

NFGNB, predominantly Pseudomonas aeruginosa and Acinetobacter baumannii, are significant healthcare-associated pathogens frequently isolated from pus and respiratory specimens. Both organisms demonstrated substantial antimicrobial resistance in this setting, with A. baumannii showing alarmingly low susceptibility to most tested agents. These findings emphasize the urgent need for continuous laboratory surveillance, formulation of local antibiograms, and stringent infection-control practices to guide empirical therapy and limit the spread of multidrug-resistant NFGNB.

DECLARATIONS

Ethics Approval: Ethical approval details were not provided in the supplied study material. The institutional ethics committee approval number and date should be inserted before submission.
Consent: The appropriate institutional policy regarding consent for use of clinical specimens and patient-related data should be stated according to the actual study protocol.
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Conflict of Interest: The authors should declare any actual conflicts of interest according to the target journal's requirements.
Author Contributions: The author contribution statement should be completed according to the actual contributions of the study team.
Data Availability: The data underlying this study are derived from microbiology laboratory records of the study institution. Data availability should be stated according to institutional policy and journal requirements.
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