Int.J.Curr.Microbiol.App.Sci (2026) *15*(7): 99-105

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 15 Number 7 (2026)**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2026.1507.010

Carbapenem Resistance among Gram-Negative Bloodstream Isolates in a Tertiary Care Hospital: A Six-Month Retrospective Analysis

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Keywords

Bloodstream infection, Carbapenem resistance, Blood culture, Antimicrobial resistance

Article Info

Received:
12 May 2026
Accepted:
22 June 2026
Available Online:
10 July 2026

A B S T R A C T

Carbapenem-resistant Gram-negative bacteria (CR-GNB) have rising as a main public health issue due to its rising incidence and limited antibiotic treatment options. Bloodstream infections caused by these strains are connected with prolonged hospitalization, and higher mortality, and increased healthcare costs. This investigation aimed to detect the presence of CR-GNB isolated from BSIs in a tertiary care hospital. This current retrospective observational investigation was done in the Department of Microbiology, Chettinad Hospital and Research Institute, Tamil Nadu, India, by collecting blood culture records from October 2023 to March 2024. A total of 1885 blood culture samples collect and processed during the investigation period, among them 425 were analysed through the BACTEC automated blood culture system. Identification of bacteria and antibiotic susceptibility test was done. Among the 425 blood cultures analysed using the BACTEC method, 196 (46.1%) were showed culture-positive, remaining 229 (53.9%) reported no growth. 29 (6.8%) GNB isolates were isolated, among them 13 (44.8%) were detected as carbapenem-resistant. The overall prevalence of carbapenem-resistant GNB was 3.1%**.** *Klebsiella pneumoniae* was the most common pathogen showed resistance to carbapenem for 76.9% (10/13, followed by *Acinetobacter baumannii* (15.4%) and *Escherichia coli* (7.7%). And interestingly none of the *Pseudomonas aeruginosa* were detected resistant to carbapenem. This investigation showed the rising incidence of CR-GNB among BSIs, with *Klebsiella pneumoniae* being the most common resistant organism. Routine antimicrobial susceptibility testing, Continuous surveillance, and effective infection prevention measures are important to decrease the carbapenem-resistant organisms spread in healthcare settings.

Introduction

Bloodstream infections (BSIs) remain the major causes of mortality and morbidity all over the world, especially among critically ill and hospitalized patients. Proper initiation of appropriate antimicrobial treatment is important for improving clinical outcomes; however, the rising prevalence of antimicrobial resistance has significantly complicating the management of BSI. Among the various resistant pathogens, the CR-GNB have rising as a major global public health issue because of their limited treatment options, increased healthcare costs, prolonged hospital stay, and high mortality rates[1,2]. Carbapenems, including Meropenem, Imipenem, doripenem, and ertapenem, are considered last-line β-lactam antibiotics for the treatment of serious infections caused by multidrug-resistant Gram-negative organisms (GNB). Unfortunately, the widespread and often inappropriate treatment of these antibiotics has rising the emergence of carbapenem resistance. The major mechanisms of resistance include the Carbapenemase enzymes production, reduced outer membrane permeability due to porin alterations, overexpression of efflux pumps, and the acquisition of mobile genetic elements carrying resistance determinants[3]. These mechanisms assist the rapid distribution of resistance among clinically significant pathogens such as *Pseudomonas aeruginosa*, *Klebsiella pneumoniae, Escherichia coli,* and *Acinetobacter baumannii*[2,4].

The World Health Organization has reported carbapenem-resistant *Acinetobacter baumannii* and carbapenem-resistant Enterobacterales as serious priority pathogens demanding urgent research and surveillance because of their considerable impact on healthcare system and patient outcomes[1]. Recent evidences have further showed the increasing burden of carbapenem-resistant BSIs, especially in tertiary care hospitals where critically ill patients are regularly exposed to invasive procedures and prolonged antibiotic treatment[2,5].

Continuous local surveillance is therefore crucial to understand the resistance, guide proper antimicrobial treatment, support antimicrobial stewardship programmes, and control practices, and strengthen infection prevention. In view of the rising threat caused by carbapenem-resistant GNB, this investigation was undertaken to identify the presence of carbapenem resistance GNB pathogens from our study population and to analyse the resistance pattern of these clinically significant pathogens.

Materials and Methods

Study design and setting: A retrospective observational investigation was done in the Department of Microbiology, Chettinad Hospital and Research Institute, Tamil Nadu, India. The investigation was carried out by reviewing blood culture results analysed from October 2023 to March 2024**.**

Study population: All blood culture samples collected in the microbiology laboratory during the study period were included for this investigation. Blood cultures yielding GNB were included for assessing carbapenem resistance. Duplicate isolates received from the same patient during the same infectious episode were excluded to avoid duplication of results. Blood culture processing and bacterial identification. Blood specimens collected from patients with suspected BSIs and were processed using the BACTEC automated blood culture system (BECTON DICKINSON (BD) BACTEC FX40) as per the manufacturer's instructions.

The BACTEC and Vitec equipment we used in this study was shown in figure 1. Blood culture bottles flagging positive were selected for preliminary test which included Gram staining, followed by subculture was done on appropriate culture media such as Chocolate agar, Blood agar, and MacConkey agar. Bacterial isolates were detected following the standard microbiological investigations such as catalase, oxidase, and handing drop method for motility detection in addition to the routinely performed biochemical tests in the clinical microbiology laboratory.

Antibiotic susceptibility testing: Antibiotic susceptibility test was performed using the Vitek 2 Compact automated ID and Antibiotic sensitivity as per routine laboratory protocol as per the CLSI-2026 guidelines applicable during the study period. Carbapenem susceptibility was detected using Vitek 2 machine, including Meropenem and Imipenem. Isolates identified as resistant to one or more carbapenem antibiotics were considered as CR-GNB.

Data collection: Demographic details, blood culture results, bacterial identification, and antibiotic susceptibility test results collected from the laboratory information system and Medical Record Department. The prevalence of carbapenem-resistant GNB was determined, and the distribution of resistant isolates among various bacterial species was detected.

Statistical analysis: The collected data were listed into Microsoft Excel and evaluated using descriptive statistics. Categorical variables were mentioned in percentages and as frequencies. The prevalence of CR-GNB was identified as the proportion of carbapenem-resistant pathogens among all blood culture samples analysed during the period and among GNB strains.

Results and Discussion

A total of 1885 blood samples was collected from the participants and analysed for this investigation. Out of 1885, 425 (22.5%) of were selected for the BACTEC method, and the remaining 1460 (77.5%) of blood cultures were selected for the conventional method.

A total of 425 blood culture samples were analysed within the study period using BACTEC method. Among these, 229 (53.9%) samples reported no bacterial growth, whereas the remaining blood samples showed positive blood culture results. Out of the culture-positive isolates samples, 29 (6.8%) were detected as GNB and were included in the investigation of carbapenem resistance. The details are shown in figure 2.

Among the 29 GNB isolates from the study samples, 13 (44.8%) were identified to be resistant to carbapenem, while the remaining 16 (55.2%) strains were sensitive to carbapenem antibiotics. When evaluated against the overall blood culture samples analysed during the period of study, the prevalence of carbapenem-resistant GNB was identified as 3.1% (13/425)**.** The details of the carbapenem resistance against the isolated GNB is shown in Figure 2

The Species-wise investigation of carbapenem-resistant strains was showed that *Klebsiella pneumoniae* was the most common pathogen, estimating for 10 (76.9%) of the 13 carbapenem-resistant strains, followed by *Acinetobacter baumannii* strains showed 2 (15.4%) strains, and *Escherichia coli* strains showed 1 (7.7%)**.** None of the isolated strains of *Pseudomonas aeruginosa* showed carbapenem-resistant during the study period. All the details are shown in figure 3.

The distribution of carbapenem-resistant strains showed that Enterobacterales members, especially *Klebsiella pneumoniae*, were the most common strains that showed carbapenem resistance among BSIs in the present investigation. The prevalence of carbapenem-resistant *K. pneumoniae* shows its rising clinical significance in tertiary care settings and highlights the need for continuous surveillance and effective antimicrobial treatment.

During the study period, a total of 425 blood samples were analysed through the BACTEC method, of which 196 were showed culture-positive samples and 29 GNB were isolated from them. Among these GNB isolated from the study population, 13 were detected as carbapenem-resistant.

In the present investigation, carbapenem-resistant GNB were detected in 13 of 425 blood culture samples, and showed the overall prevalence of 3.1%**.** Among them 44.8% (13/29) of GNB isolates showed carbapenem resistance. Though the overall prevalence among all blood cultures was low, the high number of resistance among GNB isolates showed a substantial burden of antimicrobial resistance once GNB is recognised. Similar findings have reported from tertiary care centres where carbapenem resistance remains major confined to GNB pathogens causing healthcare-associated BSIs[6,7].

Among the carbapenem-resistant strains isolated from our study population, *Klebsiella pneumoniae* were more frequently isolated for 76.9%, making it the major resistant pathogen detected in this investigation. This result is similar with several recent reports showed that carbapenem-resistant *K. pneumoniae* has become the major cause of carbapenem-resistant BSIs in many tertiary care hospitals. A prospective cohort investigation was done at Christian Medical College, Vellore, South India, showed a notable rising in carbapenem-resistant *K. pneumoniae* bacteraemia over time and reported high mortality rate even the availability of newer antimicrobial treatment[8]. Similarly, Sharma *et al.,* highlighted a large hospital outbreak caused by carbapenem-resistant *K. pneumoniae* harbouring bla_{NDM}, and bla_{OXA-48}genes, highlighting the organism's extraordinary ability to distribute within healthcare settings[9]. The prevalence of *K. pneumoniae* detected in the present investigation therefore reproduces the continuing epidemiological change towards carbapenem-resistant Enterobacterales in Indian hospitals.

The present investigation reported *Acinetobacter baumannii* as the second most common carbapenem-resistant pathogen followed by Klebsiella pneumoniae, estimating for 15.4% of resistant strains.

Figure.1 (a) BECTON DICKINSON (BD) BACTEC FX40 & (b) VITEK 2 COMPACT AUTOMATED BACTERIAL ID AND ABST SYSTEM

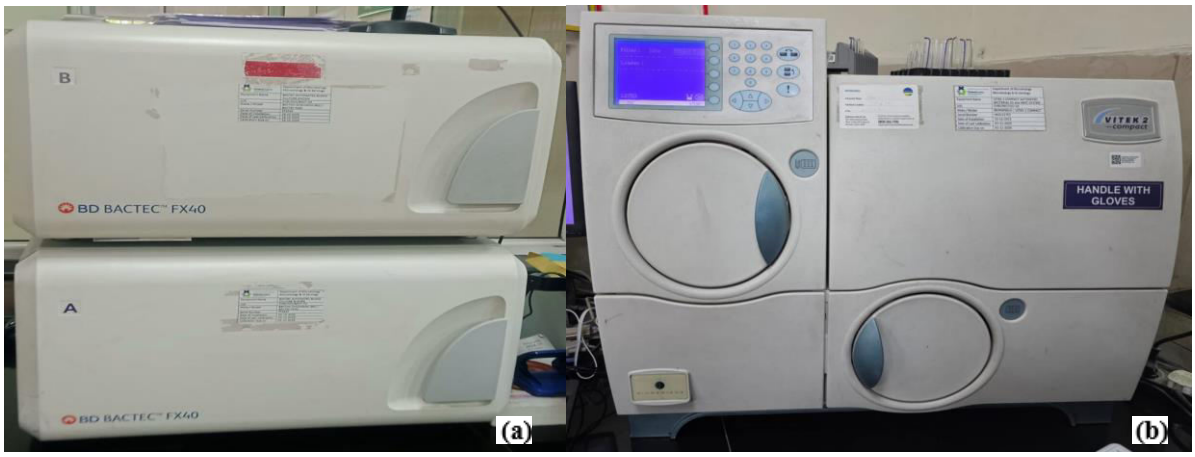


Figure.2a Distribution of blood culture analysed method among the study samples

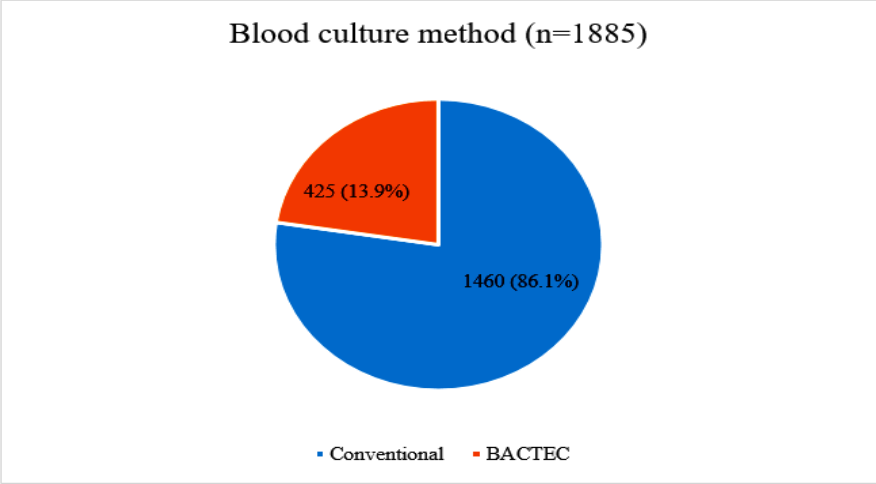


Figure.2b Distribution of carbapenem resistance strains among the GNB isolates

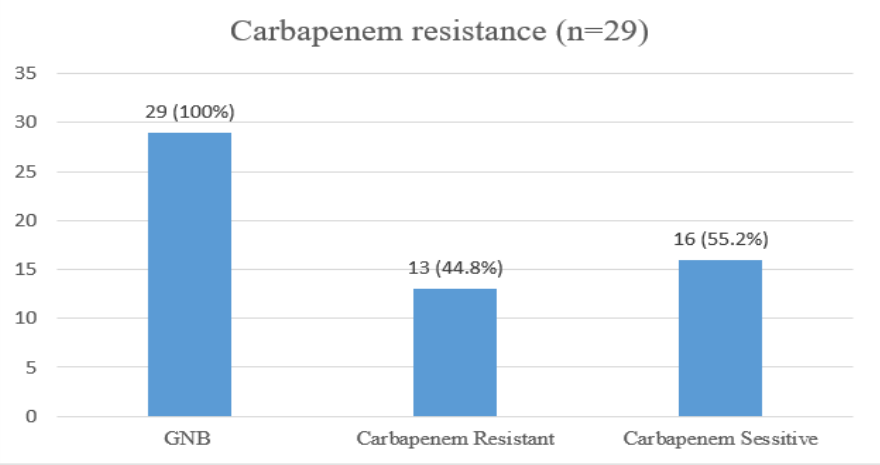
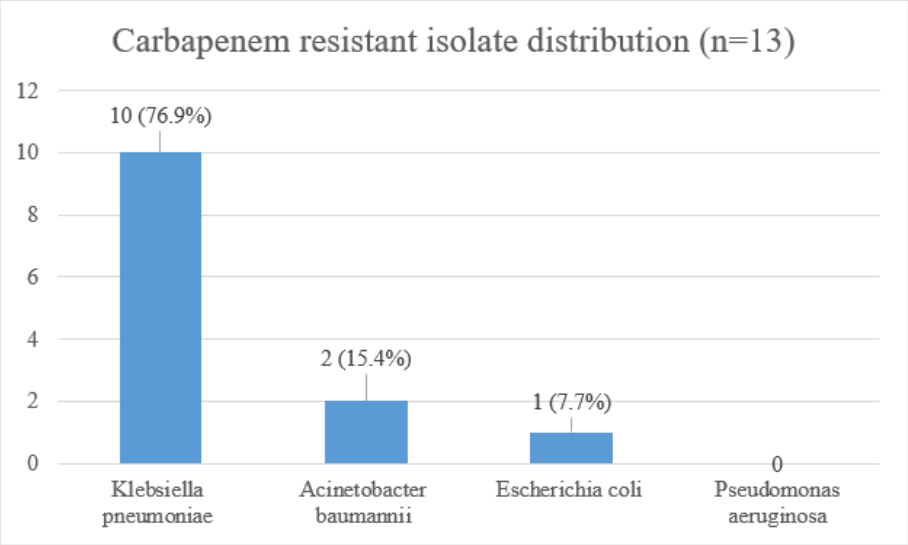


Figure.3 Species wise distribution of carbapenem resistant GNB isolates from our study population.



Even though the total number was noticeably lower than that of *K. pneumoniae*, the finding highlights the clinical significant of *A. baumannii* possesses an excellent ability to survive in hospital environments, rapidly obtain resistance factors, and cause outbreaks in intensive care units. Recent investigation reported that carbapenem-resistant *A. baumannii* as one of the most problematic multidrug-resistant strains due to its presence on environmental surfaces and limited treatment options[10,11].

From our isolated pathogen least number of isolated *Escherichia coli* showed resistance against carbapenem antibiotics, representing 7.7%**.** Carbapenem-resistant *E. coli* is usually detected less common than carbapenem-resistant *K. pneumoniae* due to Carbapenemase genes are more frequently spread among *Klebsiella* species. However, the rising emergence of carbapenem-resistant *E. coli* remains a rising concern for their important role in both the healthcare-associated infections, and community-acquired infections[8,10].

Notably, none of the isolated *Pseudomonas aeruginosa* reported resistance against carbapenem from our study population. This result contrasts with several international investigations that have reported carbapenem-resistant *P. aeruginosa* as an important factor of BSIs, especially among critically ill patients[10,11]. The absence of resistant against *P. aeruginosa* from our study isolates may be attributed to variation in study population, prevention strategies, local antimicrobial prescribing practices, or the limited number of GNB isolates included during the investigation period. Such geographical differences has been broadly reported in antimicrobial resistance surveillance investigations.

This current investigation findings further highlighting the significance of strengthening antimicrobial treatment, early laboratory findings of carbapenem-resistant strains, strict infection prevention and control measures, and routine surveillance of antimicrobial resistance. Continuous checking of resistance drifts will help clinicians in choosing appropriate empirical antimicrobial treatment while decreasing unnecessary carbapenem exposure and preventing the spread of multidrug-resistant pathogens within healthcare settings.

In conclusion, the present investigation showed that carbapenem-resistant GNB are rising among BSIs within our tertiary care hospital. While the overall incidence of carbapenem resistance among blood culture positive samples was comparatively low, nearly half of the GNB isolates showed carbapenem resistant. Notably *Klebsiella pneumoniae* existing as the major resistant pathogen. These findings highpoint the significance of continuous surveillance on antimicrobial resistance, effective infection prevention and control measures, and routine susceptibility testing to limit the rising concern and controlling the spread of carbapenem-resistant

strains in healthcare and community settings. Further multicentre investigations and molecular characterization of carbapenem resistance mechanisms are required to understand the better knowledge on transmission dynamics of these clinically significant strains.

Author Contributions

Mohan Kumar Mahadevan: Investigation, formal analysis, writing—original draft. Mohan Kumar Appadurai: Validation, methodology, writing—reviewing. Riyaz Sheriff:—Formal analysis, writing—review and editing. Priyadarshini Shanmugam: Investigation, writing—reviewing.

Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethical Approval Not applicable.

Consent to Participate Not applicable.

Consent to Publish Not applicable.

Conflict of Interest The authors declare no competing interests.

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How to cite this article:

Mohan Kumar Mahadevan, Mohan Kumar Appadurai, Riyaz Sheriff and Priyadarshini Shanmugam. 2026. Carbapenem Resistance among Gram-Negative Bloodstream Isolates in a Tertiary Care Hospital: A Six-Month Retrospective Analysis. *Int.J.Curr.Microbiol.App.Sci.* 15(7): 99-105. **doi:** https://doi.org/10.20546/ijcmas.2026.1507.010