RESEARCH PAPER

Bacteriological Profile and Antibiogram of Ventilator Associated Pneumonia: Study in a Tertiary Care Centre, South India

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ABSTRACT

Multidrug-resistant (MDR) bacterial pathogens are often the cause of ventilator-associated pneumonia (VAP), the most frequent healthcare-associated infection in intensive care unit (ICU) patients. Effective treatment depends on the prompt identification of the pathogenic organisms and their patterns of antibiotic resistance. This study was set out to assess the pattern of antibiotic susceptibility and the bacteriological spectrum in VAP cases. So, a 12-month cross-sectional observational study was conducted in the intensive care unit (ICU) of a tertiary care hospital in Vijayapura, Karnataka, South India. We collected 62 endotracheal aspirate samples from ICU patients who were mechanically ventilated for more than 48 hours and had clinical features suggestive of ventilator-associated pneumonia, including fever, leukocytosis or leukopenia, and new infiltrates on chest radiography. Quantitative culture and Gram staining were used to process the samples. We identified the bacterial isolates and assessed their susceptibility to antibiotics using the VITEK system. The results showed monomicrobial growth was found in 31 (50%) of the 62 endotracheal aspirate samples. Out of the total 31 isolates, 96.77% were Gram-negative and 3.23% Gram-positive. Klebsiella pneumoniae (41.9%) was the most frequently isolated organism, followed by Acinetobacter baumannii complex (29%). Multidrug-resistant (MDR) pathogens accounted for 26 of 31 isolates (83.87%). High antibiotic resistance rates were observed against third-generation cephalosporins (86.7%), carbapenems (80%) and fluoroquinolones (90%) among Gram-negative isolates. Colistin (96.7%) and tigecycline (100%) retained good activity against the bacterial isolates. In conclusion, the study emphasises the predominance of Gram-negative organisms in VAP, especially Klebsiella pneumoniae and Acinetobacter baumannii complex. They also exhibit high resistance to commonly used antibiotics. Agents such as tigecycline and colistin demonstrated good activity. In ICUs, monitoring of rationale use of antibiotics, stringent compliance to infection control practices, and surveillance on regular basis are critical for enhancing patient outcome and reducing resistance.

Keywords: Ventilator Associated Pneumonia, VAP, Multidrug-resistant, Klebsiella pneumoniae, Acinetobacter baumannii, Tigecycline, Colistin, ICU.

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Introduction:

Nosocomial or hospital-acquired infections remain a major cause of morbidity and mortality. Ventilator-associated pneumonia (VAP) is the second most

common infection acquired in hospitals, and it contributes significantly to hospital-acquired pneumonia cases.[1]

VAP refers to pneumonia that develops 48 hours or more after endotracheal intubation and initiation of mechanical ventilation. VAP is typically categorised as early-onset (manifesting within the initial four days of commencing mechanical ventilation) and late-onset VAP (manifesting after five or more days of commencing mechanical ventilation). Early onset VAP is typically less severe, caused mainly by bacteria which are antibiotic sensitive, and has a good prognosis. In contrast, late-onset VAP is generally linked with bacteria which are multidrug-resistant (MDR) and increased morbidity and mortality. [2, 3]

Geographical location, the duration of ICU stay, the length of mechanical ventilation, antibiotic exposure, and patient-related factors all affect the microbiological profile of ventilator-associated pneumonia.[4, 5] There has been a noticeable change in the organisms that cause VAP in recent years. To choose the most appropriate line of treatment and enhance patient outcomes, it is crucial to accurately identify the causative microorganisms and analyze their patterns of antibiotic susceptibility.[6, 7] Additionally, several recent studies have reported an increasing prevalence of MDR organisms in VAP.[8, 9] MDR bacteria continue to be a major problem for physicians because they make patient care more difficult and significantly limit the range of treatment options.[10, 11] In this context, the study was carried out to ascertain the bacteriological spectrum and the antibiotic susceptibility pattern from VAP.

Materials and Methods:

A 12-month cross-sectional observational study was conducted in the ICU of a South Indian tertiary care hospital. Sixty-two endotracheal aspirate samples were collected from patients who fulfilled the inclusion criteria, such as ICU patients on mechanical ventilation for beyond 48 hours and who displayed symptoms of ventilator-associated pneumonia (VAP), such as fever, leukocytosis or leukopenia, and new infiltrates on chest radiography. Samples exhibiting mixed bacterial growth, patients with pre-existing lung infiltrates prior to the initiation of mechanical ventilation, and patients ventilated for less than 48 hours were not included.

Endotracheal aspirate samples were collected with aseptic precautions and sent without delay to the microbiology laboratory for further processing. All samples were subjected to Gram staining followed by quantitative culture. We mechanically homogenised the samples and prepared serial dilutions using 0.9% sterile saline at 10^{-2}, 10^{-3}, and 10^{-4} concentrations. We inoculated the diluted samples on MacConkey agar (MA), Blood agar (BA) and incubated aerobically at 37ºC for 24 to 48 hours. A growth with bacterial count of $\geq 10^5$ colony-forming units per millilitre (CFU/mL) was considered significant for the diagnosis of VAP.[11]

We carried out identification of bacterial isolates and antimicrobial susceptibility testing using VITEK 2 automated system. The antibacterial agents tested include amikacin, gentamicin, aztreonam, ceftazidime, cefepime, cefoperazone/sulbactum, levofloxacin, ciprofloxacin, piperacillin/tazobactam, meropenem, imipenem, colistin, fosfomycin, tigecycline, minocycline, and trimethoprim/sulfamethoxazole. Interpreted the antimicrobial susceptibility testing (ABST) results in accordance with the CLSI guidelines.[12]

Results:

We collected endotracheal aspirates from 62 mechanically ventilated ICU patients who met the inclusion criteria during the study period from March 2024 to February 2025. Among them, 44 (71%) were males and 18 (29%) were females. The study included patients from adult, pediatric and neonatal intensive care units, with ages ranging from one month to eighty-five years. The maximum number of patients were over 60 years of age (Table 1).

TABLE 1: Age-wise distribution of VAP patients

Age group	Total (n)	Males	Females	Percentage
≤18 years	4	2	2	6.45%
19–40 years	13	7	6	20.97%
41–60 years	18	17	1	29.03%
>60 years	27	18	9	43.55%
Total	**62**	**44**	**18**	**100%**

TABLE 2: Details of number of Early-onset VAP and Late-onset VAP

Type of VAP	Number (n)	Percentage (%)
Early-onset VAP	14	22.58
Late-onset VAP	48	77.42
Total	**62**	**100**

Out of 62 VAP cases, late onset VAP (77.42%) was the most common form (Table 2). Out of 62 samples, 31

TABLE 4: Antibiotic resistance pattern of Enterobacteriaceae isolated from VAP patients

Antibiotic	K. pneumoniae (41.9%), n=13	K. oxytoca (6.5%), n=2	E. coli (6.5%), n=2	K. aerogenes (3.2%), n=1
Amoxicillin/Clavulanic acid	76.9%	100%	100%	100%
Cefuroxime	84.6%	100%	100%	100%
Cefuroxime axetil	84.6%	100%	100%	100%
Cefoperazone/Sulbactam	76.9%	50%	100%	100%
Ertapenem	69.2%	100%	0%	100%
Meropenem	76.9%	50%	100%	100%
Amikacin	69.2%	0%	100%	100%
Gentamicin	76.9%	100%	0%	100%
Ciprofloxacin	76.9%	100%	100%	100%
Tigecycline	0%	0%	0%	0%
Fosfomycin	69.2%	100%	0%	100%
Colistin	0%	0%	0%	100%
Trimethoprim/Sulfamethoxazole	84.6%	100%	50%	0%
Piperacillin/Tazobactam	84.6%	100%	0%	100%
Ceftriaxone	92.3%	100%	50%	100%
Cefepime	84.6%	100%	50%	100%
Imipenem	76.9%	50%	100%	100%

*n= number of isolates

(50%) showed positive bacterial growth. Among 31 isolates, Gram-negative bacteria were predominant, accounting for 30 (96.77%) isolates; only one isolate (3.23%) was Gram-positive i.e. methicillin-resistant *Staphylococcus aureus*. *Klebsiella pneumoniae* (n=13, 41.9%) was the most commonly isolated organism in this study, subsequently *Acinetobacter baumannii* complex (n=9, 29%), *Pseudomonas aeruginosa* (n=3, 9.7%), *Klebsiella oxytoca* and *Escherichia coli* (n=2 each, 6.5%), and *Klebsiella aerogenes* (n=1, 3.2%).

TABLE 3: Details of number of isolates

Name of the organism isolated	Number of isolates (n)	Percentage (%)
Klebsiella pneumoniae	13	41.94
Acinetobacter baumannii complex	9	29.03
Pseudomonas aeruginosa	3	9.68
Klebsiella oxytoca	2	6.45
Escherichia coli	2	6.45
Klebsiella aerogenes	1	3.23
Methicillin-resistant *Staphylococcus aureus* (MRSA)	1	3.23
Total isolates	**31**	**100**

TABLE 5: Antibiotic resistance pattern of non-fermenters isolated from VAP patients

Antibiotic	A. baumannii complex (29%), n=9	P. aeruginosa (9.7%), n=3
Amoxicillin/Clavulanic acid	0%	—
Cefuroxime	0%	—
Cefuroxime axetil	0%	—
Cefoperazone/Sulbactam	55.6%	66.7%
Ertapenem	0%	100%
Meropenem	100%	66.7%
Amikacin	88.9%	33.3%
Gentamicin	100%	0%
Ciprofloxacin	100%	100%
Tigecycline	0%	0%
Fosfomycin	100%	—
Colistin	0%	0%
Trimethoprim/Sulfamethoxazole	66.7%	0%

Antibiotic	A. baumannii complex (29%), n=9	P. aeruginosa (9.7%), n=3
Piperacillin/Tazobactam	100%	100%
Ceftriaxone	100%	—
Cefepime	77.8%	33.3%
Imipenem	100%	66.7%
Ceftazidime	—	33.3%
Levofloxacin	—	100%

*n= number of isolates

Among the Gram-negative isolates, resistance to carbapenems was observed in 80% of isolates, while resistance to third-generation cephalosporins and quinolones was noted in 86.7% and 90% of isolates, respectively, indicating a high prevalence of multidrug resistance. Among *Klebsiella pneumoniae* isolates, resistance was observed to ceftriaxone in 92.3%, cefuroxime in 84.6%, meropenem and imipenem in 76.9%, and ciprofloxacin in 76.9% of strains. All *Acinetobacter baumannii* complex isolates (100%) were resistant to carbapenems and showed poor susceptibility to amikacin, gentamicin, and fluoroquinolones. Among *Pseudomonas aeruginosa* isolates, 66.7% exhibited multidrug resistance.

Tigecycline showed excellent susceptibility among Enterobacterales, with 92-100% susceptibility, and moderate activity against *A.baumannii* (66.7% intermediate susceptibility). All Gram-negative isolates were found susceptible to colistin using VITEK 2 method. (Table 4 and Table 5).

The single MRSA isolate was resistant to ciprofloxacin, benzylpenicillin, erythromycin, and oxacillin, but sensitive to daptomycin, vancomycin, linezolid, teicoplanin, rifampicin, tetracycline and tigecycline.

Multidrug Resistance (MDR) was observed in 26 of 31 isolates (83.87%), particularly among *K. pneumoniae* and *Acinetobacter* spp., suggesting a high likelihood of extended-spectrum β-lactamase (ESBL), AmpC β-lactamase (AmpC), and metallo β-lactamase (MBL) producing strains.

Discussion:

In this study, the bacterial spectrum and antimicrobial susceptibility patterns of VAP pathogens in the ICUs were investigated. To improve clinical outcomes and guide suitable empirical therapy, it is essential to comprehend regional bacterial prevalence and resistance patterns. The findings of this study demonstrate the increasing threat posed by MDR Gram-negative bacteria in extremely ill patients and offer baseline data to support antimicrobial stewardship strategies.

In the current study, a male predominance was observed in patients with VAP, with 44 males (70.97%) and 18 females (29.03%). This gender distribution is comparable to the outcomes of Patro et al. [4] and Gupta et al. [7], where males constituted the majority of VAP cases.

In the current study, the majority of ventilator-associated pneumonia cases occurred in patients aged more than 60 years (43.55%), followed by those aged 41–60 years (29.03%), indicating a clear predominance of VAP among older adults. Similar age distributions have been reported by Patro et al. [4], who observed a higher incidence of VAP in patients above 50 years, and Gupta et al. [7], where most cases belonged to the older age groups. Advancing age is a well-recognized risk factor for VAP due to waning immune function, occurrence of numerous comorbidities, prolonged ICU stay, and increased need for mechanical ventilation.[13, 14] Late-onset VAP predominated in the current study, which is comparable with earlier findings from Somi Patro et al. [4], who reported 70% of cases, and Vijeta Sharma et al. [8], who found a late-onset VAP prevalence of 68%. Early-onset VAP, on the other hand, was less common in these studies, highlighting the fact that late-onset VAP is more prevalent in intensive care unit patients and linked to MDR Gram-negative bacilli, which can lead to poorer clinical outcomes and fewer treatment options.[15] Additional risk factors that may predispose patients to VAP include prolonged ICU stay, underlying comorbidities, invasive procedures, and impaired host immunity.[4]

In contrast to the 88.5% reported by Gupta et al., our study's culture positive rate was just 50%.[7] Previous antibiotic exposure in a significant portion of our study population might be responsible for this discrepancy. Compared to the rates reported by Patro et al.[4] (85.1%) and Gupta et al.[7] (88.3%) studies, Gram-negative organisms accounted for 96.77% of culture positive cases in the current investigation. This observation supports the consistent patterns found in ICU-based research and highlights the notable preponderance of Gram-negative bacilli in VAP.

Cross-transmission within intensive care units is facilitated by gram-negative bacteria's greater ability to survive in damp hospital settings, such as ventilator circuits, humidifiers, and sinks. They are even more resistant and dominant in the intensive care unit due to their innate and acquired resistance mechanisms.[16, 17]

Klebsiella pneumoniae was the most commonly isolated organism (41.94%) among the Gram-negative pathogens. This is in contrast to the results of Gupta et al.[7], where *Pseudomonas aeruginosa* was the most prevalent isolation (38.2%), and Patro et al.[4], who reported *Acinetobacter spp.* as the commonest isolate (21.27%). These variations point to a growing prevalence of *Klebsiella pneumoniae* as a primary cause of VAP in our ICU.

The high prevalence of MDR Gram-negative pathogens in the present study has important clinical implications. Infections caused by these organisms are associated with prolonged ICU stay, increased healthcare costs, limited therapeutic options, and increased mortality.[4] The predominance of carbapenem-resistant *Klebsiella pneumoniae* and *Acinetobacter baumannii* complex in our ICU setting reflects the growing burden of antimicrobial resistance across tertiary care centres in India.

The observed variation in the predominant pathogen across studies may be attributed to differences in local ICU ecology, antibiotic prescribing practices, and infection control measures[18]. Increased use of broad-spectrum cephalosporins and carbapenems may selectively favour the emergence and persistence of *Klebsiella pneumoinae*, particularly carbapenem-resistant strains.[19, 20]

The antimicrobial resistance patterns observed in the present study are comparable with reports from other Indian ICUs, where increasing resistance to cephalosporins, carbapenems, and fluoroquinolones has been documented. Excessive use of broad-spectrum antibiotics, prolonged hospitilization, invasive procedures, and cross-transmission within ICUs may contribute to this trend. These findings highlight the urgent need for strict infection prevention control practices, implementation of antimicrobial stewardship programmes, and periodic surveillance of local antibiograms to guide appropriate empirical therapy.

Antimicrobial resistance patterns showed both similarities and variations across studies. In the study by Patro et al.[4] *Klebsiella pneumoniae* demonstrated 100% resistance to Ceftazidime, Cefotaxime and Ciprofloxacin. In comparison, our study also revealed high resistance rates, particularly to Ceftriaxone (92.31%), Cefuroxime (84.62%), and Ciprofloxacin (84.62%). Taken together, these findings are consistent with the growing trend of increasing anti-microbial resistance among *Klebsiella pneumoniae* isolates.

Carbapenem resistance among *Klebsiella pneumoniae* was also high in our study, with 76.92% isolates resistant to meropenem. Similarly, 75% carbapenem resistance was reported by Patro et al.[4] suggesting wide spread circulation of carbapenem-resistant *Klebsiella pneumoniae* strains.

Acinetobacter baumannii complex was the second most common isolates (29.03%) in our study. All isolates were resistant to meropenem (100%), these results align with those stated by Sharma et al.,[8] who demonstrated that carbapenem resistance in *Acinetobacter* isolates was also universal. These data show the critical issue of multidrug resistance in *Acinetobacter* species across Indian ICUs.

Tigecycline and colistin retained relatively good activity against most Gram-negative isolates of these studies[7, 8] as well as in our study. These findings suggest that certain antibiotics continue to have value as targeted therapy for MDR VAP pathogens.

Pseudomonas aeruginosa constituted 9.68% of the isolates in our study, which is lower than the 38.2% reported by Gupta et al.[7] In our isolates, 66.7% were multidrug resistant (MDR), compared to 73.1% MDR reported in Gupta's study. While prevalence varied, the MDR rates remained comparably high.

In our study, we isolated MRSA in 3.23% of VAP cases, which is lower than the rates stated by Patro et al. [4] and Gupta et al.[7], who observed 8% and 12% MRSA isolates respectively.

Limitations:

Limitation of this study is that samples showing mixed bacterial growth were excluded, which may have led to an underestimation of the true burden of polymicrobial VAP.

Conclusion:

This study demonstrates a substantial burden of MDR Gram-negative organisms causing VAP in a tertiary care ICU setting in South India, with *Klebsiella pneumoniae* and *Acinetobacter baumannii* complex being the predominant pathogens. High resistance rates to commonly used antibiotics, including β-lactam / β-lactamase inhibitor combinations, fluoroquinolones, carbapenems, and third-generation cephalosporin limit therapeutic options

Knowledge of local antimicrobial susceptibility patterns is essential for guiding empirical therapy and improving clinical outcomes. In our setting, agents such as tigecycline, polymyxins, and vancomycin retained better activity against MDR pathogens and may be considered for targeted treatment.

The findings of the present study emphasize the urgent need for continuous microbiological surveillance, institution-specific antibiograms, strict infection prevention and control practices, and antimicrobial stewardship programmes to reduce the emergence and spread of antimicrobial resistance within ICUs.

Declarations

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Conflict of Interest

The authors declare that there is no conflict of interest regarding the publication of this manuscript.

Authors Contribution

Dr Ashok conceived and designed the study, performed microbiological processing and data collection, analysed the data, and drafted the manuscript. Dr Smitha Bagali contributed to study supervision, interpretation of findings, and critically reviewed the manuscript for important intellectual content. Dr. Rashmi Karigoudar contributed to study design, data interpretation, and critical revision of the manuscript. Dr. Annapurna Sajjan contributed to interpretation of findings and manuscript review. Dr Praveen R Shahapur contributed to supervision, interpretation of results, and critical revision of the manuscript. Dr Laxmi Kakhandki contributed to manuscript review and provided intellectual input during manuscript preparation. All authors reviewed the manuscript, approved the final version, and agreed to be accountable for all aspects of the work.

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Ethics statement

This study was approved by Institutional Ethics Committee of Shri B M Patil Medical College Hospital and Research Institute.

Informed Consent

Written informed consent was obtained from all participants or their legally authorized representatives prior to inclusion in the study. Patient confidentiality and privacy were strictly maintained throughout the study.

Data Availability

All data generated and analyzed during this study are included in this published article. Additional data supporting the findings of this study are available from the corresponding author upon reasonable request.

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