



Antimicrobial resistance, ESBLs and Carbapenemase producing *E. Coli* and *Klebsiella pneumoniae* isolated from positive blood culture samples in Tertiary Care Hospitals Delhi NCR

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ABSTRACT:

Bacterial antimicrobial resistance (AMR), which arises when changes in bacteria make the drugs used to treat infections less effective, has become one of the major public health threats of the 21st century. Strains of Enterobacteriaceae that produce extended spectrum beta-(β)-lactamase (ESBL) have now become a significant issue among hospitalized patients. Since the 1980s, a dramatic rise in the number of β -lactamases has been reported of the last century, but this rise is attributable almost entirely to class A and D β -lactamases. Among the class A β -lactamases, the extended-spectrum β -lactamases (ESBLs), which can hydrolyze expanded-spectrum cephalosporins (such as cefotaxime, ceftriaxone, cefatazidime, or cefepime) and monobactams (aztreonam), constitute a public health concern. Class A ESBLs primarily comprise TEM, SHV, CTX-M, VEB, and GES enzymes. Of these, the largest number of variants reported in recent years belongs to the CTX-M. This explosive spread of CTX-Ms across the globe has been termed the “CTX-M pandemic” because of their growing description worldwide. Although various updates on CTX-M β -lactamases have been published, new information regarding the dispersion and clonality of CTX-M producing isolates, molecular epidemiology, protein plasticity, evolution and origin of the blaCTX-M genes, the influence of antibiotic use, and patients risk factors, justify this new review. Carbapenems are a class of antibiotics frequently reserved as a last-resort treatment for multidrug-resistant (MDR) Gram-negative bacterial infections. The carbapenemase genes that are reported most often consist of blaNDM-1, blaVIM, blaIMP, bla OXA-48, bla OXA-181, and bla KPC. This is a retrospective, a cohort study involving patients with bacterial bloodstream infection who were admitted to our multi centre tertiary care hospitals Delhi- NCR from January 1, 2023, through December 31, 2025. A total of 41,871 samples were collected during the study period, of which 5,858 (14.0%) were culture positive samples. *Escherichia coli* was the most frequently isolated organism, accounting for 3,356 cases (57.28%). *Klebsiella pneumoniae* was the isolated. Resistance was highest to cefuroxime/cefuroxime axetil (89.85%), followed by ceftriaxone (88.59%), nalidixic acid (84.82%), ciprofloxacin (83.52%), and levofloxacin (78.91%).

Keyword: ESBL, Carbapenemase, Antimicrobial Resistant, Blood culture samples

INTRODUCTION

Bacterial antimicrobial resistance (AMR), which arises when changes in bacteria make the drugs used to treat infections less effective, has become one of the major public health threats of the 21st century. The Review on Antimicrobial Resistance, commissioned by the UK Government, contended that AMR could cause 10 million deaths annually by 2050 (Murray C JL et al., 2019)

Infections are frequently caused by Gram-negative bacteria (GNB), and the organisms most often isolated in bloodstream infection (BSI) cases are *Escherichia coli*, *Klebsiella pneumoniae*, and other Enterobacteriales (previously reported as Enterobacteriaceae). Among many GNB organisms, the upregulation or acquisition of antimicrobial resistance mechanisms is extremely efficient, particularly when antimicrobial selection pressure is present. (De Angelis G et al., 2020). Infections of the bloodstream (BSIs) have a major effect on health and economies around the world. In 2017, roughly 49 million sepsis cases occurred globally, resulting in 11 million deaths, and 41% of those deaths were among children under 5. India faces a particularly heavy burden, with an estimated 11 million cases and 3 million deaths. A recent study revealed that sepsis was involved in 56% of cases across 35 Indian ICUs, and 45% of those were linked to multi-drug resistant organisms (Kaur J et al., 2024). Strains of Enterobacteriaceae that produce extended spectrum beta-(β)-lactamase (ESBL) have now become a significant issue among hospitalized patients. These organisms cause many different infections, including septicemia, urinary tract infection, hospital-acquired pneumonia, intra-abdominal abscess, brain abscess, and device-related infections (Taneja j et al 2010). Since the 1980s, a dramatic rise in the number of β -lactamases has been reported of the last century, but this rise is attributable almost entirely to class A and D β -lactamases. Among the class A β -lactamases, the extended-spectrum β -lactamases (ESBLs), which can hydrolyze expanded-spectrum cephalosporins (such as cefotaxime, ceftriaxone, cefatazidime, or cefepime) and monobactams (aztreonam), constitute a public health concern. Class A ESBLs primarily comprise TEM, SHV, CTX-M, VEB, and GES enzymes. Of these, the largest number of variants reported in recent years belongs to the CTX-M family (123 variants up to 2011, last accession December 5, 2011) 1. This explosive spread of CTX-Ms across the globe has been termed the “CTX-M pandemic” because of their growing description worldwide. Although various updates on CTX-M β -

lactamases have been published, new information regarding the dispersion and clonality of CTX-M producing isolates, molecular epidemiology, protein plasticity, evolution and origin of the blaCTX-M genes, the influence of antibiotic use, and patients risk factors, justify this new review. (Cantón R et al., 2012). Carbapenems are a class of antibiotics frequently reserved as a last-resort treatment for multidrug-resistant (MDR) Gram-negative bacterial infections. Bacteria categorized as Carbapenemase-producing Enterobacterales (CPE) possess the capacity to break down carbapenems, thus escaping the antimicrobial effects of these drugs. Given how crucial carbapenems are for combating MDR bacterial infections, the dissemination of CPE constitutes a significant global concern, further narrowing the therapeutic alternatives at hand. (Alvisi G et al., 2025). The carbapenemase genes that are reported most often consist of blaNDM-1, blaVIM, blaIMP, bla OXA-48, bla OXA-181, and bla KPC. (Khan Z et al., 2025). One study demonstrated that each hour of delay in giving effective antimicrobial treatment lowered survival rates by 9 %; a separate study found that even when active antimicrobial therapy for bacteremia caused by CRE infection was started within 47 h, sepsis due to CRE organisms led to a 49 % mortality rate within 30 days (Hoj TR et al., 2021).

This study, is intended to assess the bacterial pathogens identified from positive blood cultures and to establish their antimicrobial resistance patterns. The research will offer epidemiological data on the distribution of bacterial isolates and their resistance profiles, MDR, XDR, PDR, ESLBs and CRE within the chosen tertiary-care hospitals. It is anticipated that these findings will enhance knowledge of local bloodstream infection epidemiology, assist in evidence-based selection of antimicrobials, and supply valuable information for antimicrobial stewardship, infection prevention, and upcoming surveillance of antimicrobial resistance across the Delhi NCR region.

MATERIALS AND METHODS:

Study design and Data Collection

This is a retrospective, a cohort study involving patients with bacterial bloodstream infection who were admitted to our multi centre tertiary care hospitals Delhi- NCR from January 1, 2023, through December 31, 2025. The study was approved by the local Institutional Ethics Committee. The hospitals come patients both directly from the local community and referred from regional hospitals. All positive blood sample that tested positive for cultures containing identified bacterial pathogens from individuals, who had been admitted to the hospital over the course of the study were enrolled in the study. The standard microbiological profile was documented for every clinical sample that arrived at the lab.

Blood samples were processed in BacT/Alert (BioMérieux, France) bottles upon clinical suspicion of BSI. Bottles that signaled positive were then subculture on blood, MacConkey, and Sheep Blood agar. These plates were incubated aerobically at 37°C and examined after 18-24 hours. Bacterial identification was done by the Vitek 2 Compact system (BioMérieux, France). Antimicrobial susceptibility was done by Vitek 2 Compact system (BioMérieux, France) and reported as per Clinical and Laboratory Standards Institute (CLSI) guidelines.

For Gram-negative isolates, MIC testing was using Vitek 2 compact system (BioMérieux, France) performed for the following antimicrobials for each isolate: The antibiotics included are cefuroxime, cefuroxime axetil, ceftriaxone, nalidixic acid, ciprofloxacin, levofloxacin, aztreonam, cefepime, trimethoprim/sulfamethoxazole, ceftazidime, amoxicillin/clavulanic acid, piperacillin/tazobactam, ticarcillin/clavulanic acid, ertapenem, meropenem, cefoperazone/sulbactam, gentamicin, imipenem, doripenem, tigecycline, minocycline, amikacin, and colistin. Antibiotic susceptibilities were performed using CLSI guidelines with breakpoints from 2025. [CLSI 2025]

Data entry and statistical analysis

Transfer data from a VITEK 2 Compact system to Microsoft Excel and analyzed using Statandard

Percentage comparison was done by the Chi-square test. Susceptibility pattern was analyzed using Spearman's correlation test. Significance of statistical difference was considered less than 0.05.

RESULTS

Patient Demographics

A total of 41,871 samples were collected during the study period, of which 5,858 (14.0%) were culture positive and included in the analysis. Among the culture-positive cases, 3,648 (62.27%) were male and 2,210 (37.72%) were female, (Figure 1) with a male-to-female ratio of 1.65:1.

Among male patients, *Escherichia coli* was isolated in 1,968 cases (58.64%), while *Klebsiella pneumoniae* was identified in 1,680 cases (67.15%). In female patients, *Escherichia coli* was isolated in 1,388 cases (41.36%), whereas *Klebsiella pneumoniae* was detected in 822 cases (32.85%). A total of 5858 positive blood culture isolates of *Escherichia coli* and *Klebsiella pneumoniae* were included for bacterial identification, antimicrobial susceptibility testing, and phenotypic detection of ESBL and carbapenemase production.

Prevalence of Escherichia coli and Klebsiella pneumoniae

Escherichia coli was the most frequently isolated organism, accounting for 3,356 cases (57.28%). *Klebsiella pneumoniae* was the isolated organism,

accounting for 2,502 cases (42.71%). *Escherichia coli* showed a higher prevalence compared to *Klebsiella pneumoniae* among culture-positive isolates. This indicates the predominance of *E. coli* in the study.

Overall Antibiotic resistance pattern Escherichia coli and Klebsiella pneumoniae

Resistance was highest to cefuroxime/cefuroxime axetil (89.85%), followed by ceftioxone (88.59%), nalidixic acid (84.82%), ciprofloxacin (83.52%), and levofloxacin (78.91%). High resistance was also observed to aztreonam (73.64%), cefepime (70.04%), trimethoprim/sulfamethoxazole (67.81%), amoxicillin/clavulanic acid (66.20%), and piperacillin/tazobactam (63.15%). Moderate resistance was noted to ticarcillin/clavulanic acid (54.46%), ertapenem (53.77%), cefoperazone/sulbactam (48.43%), meropenem (49.29%), gentamicin (46.22%), and imipenem (46.00%). Resistance was comparatively lower to colistin (7.44%), tigecycline (29.15%), amikacin (22.79%), and minocycline (28.08%). (Table 1)

Antimicrobial susceptibility profile of Klebsiella pneumoniae

The antimicrobial susceptibility profile of *Klebsiella pneumoniae* showed extensive resistance across most antibiotic classes. The highest resistance was observed against cefuroxime/cefuroxime axetil (86.07%), ceftioxone (84.04%), aztreonam (77.96%), and ciprofloxacin (76.08%). Carbapenem resistance was also substantial, ranging from 57.38% to 63.95%. Colistin showed the highest susceptibility (81.04%), followed by tigecycline, amikacin, and minocycline. Overall, the findings indicate a high prevalence of multidrug-resistant *K. pneumoniae* with limited effective antibiotic options. (Table 2)

Antimicrobial susceptibility profile of Escherichia coli

The antimicrobial susceptibility profile of *Escherichia coli* showed high susceptibility to colistin (94.84%), tigecycline (87.29%), amikacin (77.74%), and minocycline (73.13%). Carbapenems showed moderate activity, with doripenem being the most active (71.43%). In contrast, very high resistance was observed to nalidixic acid (93.69%), cefuroxime/cefuroxime axetil (92.62%), ceftioxone (91.91%), ciprofloxacin (89.06%), and levofloxacin (86.16%). Overall, the findings indicate substantial multidrug resistance. (Table 3)

Multidrug Resistant (MDR) and Extensively Drug-Resistant (XDR) and Pan Drug-Resistant (PDR) Isolates

Escherichia coli had a higher prevalence of MDR (28.04%), whereas *Klebsiella pneumoniae* exhibited a higher prevalence of XDR (55.64%). PDR was observed at low levels in both organisms *Escherichia coli* 0.83% and *Klebsiella pneumoniae* 1.60%, respectively.

Multidrug Resistant (MDR) Isolates

Multidrug-resistant (MDR) isolates among *Escherichia coli* and *Klebsiella pneumoniae*. Among the two organisms, *Escherichia coli* exhibited a higher prevalence of MDR isolates (28.04%) compared to *Klebsiella pneumoniae* (11.31%). These findings indicate that multidrug resistance was more frequently observed in *E. coli* than in *K. pneumoniae* in the present study.

Extensively Drug Resistant (XDR) Isolates

Extensively drug resistant (XDR) isolates among *Escherichia coli* and *Klebsiella pneumoniae*. The prevalence of XDR was 9.89% among *Escherichia coli* isolates, whereas *Klebsiella pneumoniae* exhibited a markedly higher prevalence of 55.64%. These findings indicate that XDR was substantially more common in *K. pneumoniae* than in *E. coli* in the present study.

Pan Drug Resistant (PDR) Isolates

Pan drug resistant (PDR) isolates among *Escherichia coli* and *Klebsiella pneumoniae*. The prevalence of PDR was 0.83% among *Escherichia coli* isolates and 1.60% among *Klebsiella pneumoniae* isolates. Although the prevalence of PDR was low for both organisms, *K. pneumoniae* exhibited a slightly higher proportion of PDR isolates than *E. coli*. The detection of PDR isolates is of significant clinical concern because these organisms exhibit resistance to all tested antimicrobial agents, leaving very limited or no effective treatment options.

Extended-Spectrum β -Lactamase (ESBL) Production Isolates

ESBL Producing *Escherichia coli*

Among the two organisms, *Escherichia coli* demonstrated a higher prevalence of ESBL production, with 46.36% of isolates identified as ESBL producers. In comparison, 22.34% of *Klebsiella pneumoniae* isolates were found to produce ESBL.

ESBL Producing *Klebsiella pneumoniae*

The present study demonstrated that 22.34% of *Klebsiella pneumoniae* isolates were identified as extended-spectrum β -lactamase (ESBL) producers. This finding indicates that approximately one-fifth of the *K. pneumoniae* isolates were capable of producing ESBL enzymes, which confer resistance to extended-spectrum β -lactam antibiotics, particularly third-generation cephalosporins and monobactams. The presence of ESBL-producing *K. pneumoniae* represents a significant concern because these isolates are commonly associated with multidrug resistance, thereby limiting the effectiveness of commonly prescribed antimicrobial agents.

Carbapenemase production Isolates

Carbapenemase Producing *Escherichia coli*

The present study demonstrated that 31.82% of *Escherichia coli* isolates were carbapenem-resistant. This finding indicates that nearly one-third of the *E. coli* isolates exhibited resistance to carbapenems, which are considered last-line antibiotics for the treatment of severe infections caused by multidrug-resistant Gram-negative bacteria. Carbapenem resistance is commonly associated with the production of carbapenemase enzymes, although other resistance mechanisms such as porin loss combined with β -lactamase production or efflux pump overexpression may also contribute.

Carbapenemase producing *Klebsiella pneumoniae*

The present study demonstrated that 43.84% of *Klebsiella pneumoniae* isolates were carbapenem-resistant. This finding indicates that a substantial proportion of *K. pneumoniae* isolates exhibited resistance to carbapenems, which are considered last-resort antibiotics for the treatment of severe infections caused by multidrug-resistant Gram-negative bacteria.

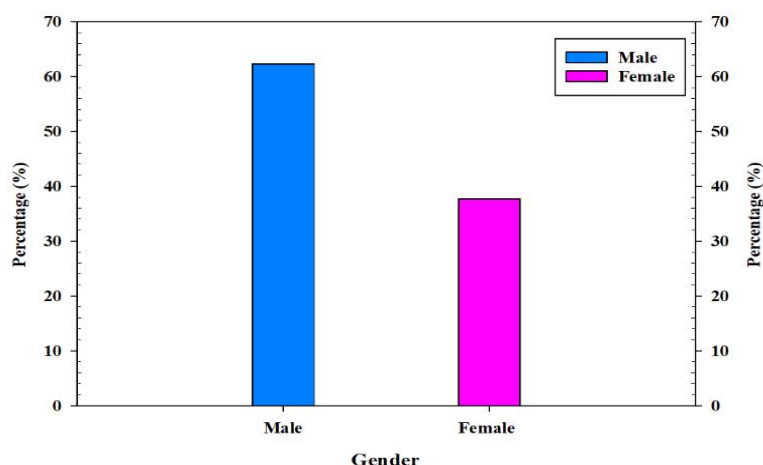


Table No. 1 Overall Antibiotic resistance pattern *Escherichia coli* and *Klebsiella pneumoniae* isolated from Positive blood culture samples

Antibiotic agents	Resistant (n) %	
Cefuroxime	(4240)	89.85
Cefuroxime Axetil	(4240)	89.85
Ceftriaxone	(4176)	88.59
Nalidixic Acid	(715)	84.82
Ciprofloxacin	(4859)	83.52
Levofloxacin	(872)	78.91
Aztreonam	(813)	73.64
Cefepime	(4071)	70.04
Trimethoprim/Sulfamethoxazole	(3944)	67.81
Ceftazidime	(742)	67.27
Amoxicillin/Clavulanic Acid	(3124)	66.20
Piperacillin/Tazobactam	(3668)	63.15
Ticarcillin/Clavulanic Acid	(116)	54.46
Ertapenem	(2536)	53.77
Meropenem	(2869)	49.29
Cefoperazone/Sulbactam	(2820)	48.43
Gentamicin	(2689)	46.22
Imipenem	(2673)	46.00
Doripenem	(87)	41.04
Tigecycline	(1696)	29.15
Minocycline	(310)	28.08
Amikacin	(1325)	22.79

Colistin	433	7.44
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Table No. 2 Antimicrobial susceptibility profile of *Klebsiella pneumonia* isolated from Positive blood culture samples

Antibiotic agents	Sensitivity (n) %	
Colistin	2013	81.03
Tigecycline	1211	48.77
Amikacin	1195	48.14
Minocycline	225	45.91
Gentamicin	1087	43.79
Cefoperazone/Sulbactam	986	39.66
Meropenem	983	39.54
Trimethoprim/Sulfamethoxazole	958	38.61
Imipenem	905	36.49
Ertapenem	714	35.82
Doripenem	30	34.88
Piperacillin/Tazobactam	752	30.33
Nalidixic Acid	94	29.37
Ticarcillin/Clavulanic Acid	22	25.58
Cefepime	633	25.51
Amoxicillin/Clavulanic Acid	487	24.39
Aztreonam	105	21.42
Ceftazidime	99	20.20
Levofloxacin	82	16.70
-Ceftriaxone	313	15.70
Ciprofloxacin	384	15.46
Cefuroxime	232	11.62
Cefuroxime Axetil	232	11.62

Table No. 3 Antimicrobial susceptibility profile of *Escherichia coli* isolated from Positive blood culture samples

Antibiotics	Sensitive (n) %	
Colistin	3164	94.84
Tigecycline	2911	87.29
Amikacin	2591	77.74
Minocycline	449	73.13
Doripenem	90	71.43
Imipenem	2026	60.82
Gentamicin	1978	59.29
Meropenem	1916	57.45
Ertapenem	1368	50.24
Cefoperazone/Sulbactam	1605	48.10
Ticarcillin/Clavulanic Acid	55	43.31
Piperacillin/Tazobactam	1212	36.41
Aztreonam	177	28.83
Trimethoprim/Sulfamethoxazole	914	27.41
Amoxicillin/Clavulanic Acid	691	25.38
Ceftazidime	129	21.04
Cefepime	647	19.42
Ceftriaxone	215	7.90
Nalidixic Acid	33	6.31
Cefuroxime	146	5.36
Cefuroxime Axetil	146	5.36

Levofloxacin	28	4.56
Ciprofloxacin	149	4.47

DISCUSSION

In this present study, 41,871 samples were collected during the study period, of which 5,858 (14.0%) were culture positive. Among the culture-positive cases, 3,648 (62.27%) were male and 2,210 (37.72%) were female. Among male patients, *Escherichia coli* was isolated in 1,968 cases (58.64%), while *Klebsiella pneumoniae* was identified in 1,680 cases (67.15%). In female patients, *Escherichia coli* was isolated in 1,388 cases (41.36%), whereas *Klebsiella pneumoniae* was detected in 822 cases (32.85%). In this study, *E. coli* was the most common isolate, representing 3,356 (57.28%) cases, while *K. pneumoniae* came second with 2,502 (42.71%) cases. By contrast, Datta et al. found *K. pneumoniae* in 21.9% (1,271) and *E. coli* in 19.06% (1,103) of the Gram-negative isolates. The greater prevalence of *E. coli* observed in this study could be explained by variations in the study population, hospital setting. (Datta S et al. 2022)

In the present study, resistance was highest to cefuroxime/cefuroxime axetil (89.85%), followed by ceftriaxone (88.59%), nalidixic acid (84.82%), ciprofloxacin (83.52%), and levofloxacin (78.91%). Similarly, Dr. Gaurishanker Shrimali et al. reported high resistance to beta-lactam antibiotics, including ampicillin, cefotaxime, and ceftazidime, with more than 70% of *E. coli* and 80% of *K. pneumoniae* isolates showing non-susceptibility. High resistance to ciprofloxacin and levofloxacin was also observed, which is consistent with the present findings. (Dr. Gaurishanker Shrimali, et al., 2025)

In the present study, *E. coli* showed high susceptibility to colistin (94.84%), tigecycline (87.29%), amikacin (77.74%), and minocycline (73.13%), whereas *K. pneumoniae* showed high resistance to cefuroxime/cefuroxime axetil (86.07%), ceftriaxone (84.04%), aztreonam (77.96%), and ciprofloxacin (76.08%). In comparison, the other study reported 100% susceptibility of *E. coli* to imipenem, meropenem, ertapenem, and tigecycline, while *K. pneumoniae* showed 95.1% susceptibility to these carbapenems and 100% to tigecycline. However, susceptibility to amikacin was lower in *K. pneumoniae* (36.6%) and *E. coli* (70%). These findings indicate considerable variation in antimicrobial susceptibility between studies, possibly due to differences in study population, antibiotic exposure, and local resistance patterns. (Taneja et al., 2010). *Escherichia coli* had a higher prevalence of MDR (28.04%), whereas *Klebsiella pneumoniae* exhibited a higher prevalence of XDR (55.64%). PDR was observed at low levels in both organisms *Escherichia coli* 0.83% and *Klebsiella pneumoniae* 1.60%, respectively. In the present study, ESBL production was higher in *E. coli* (46.36%) than in *K. pneumoniae* (22.34%). Similarly, Taşkın Kafa et al. reported higher ESBL positivity in *E. coli* (41.2%) than in *K. pneumoniae* (25%). In contrast, Dr. Gaurishanker Shrimali et al. reported ESBL production of 58% in *E. coli* and 64% in *K. pneumoniae*. The variation in ESBL rates may be due to differences in study population, geographic location, antibiotic exposure, and laboratory methods. [(Kafa et al., 2023) (Dr. Gaurishanker Shrimali, et al., 2025)]

In the present study, carbapenem resistance was observed in 31.82% of *E. coli* and 43.84% of *K. pneumoniae* isolates, while carbapenemase production was confirmed in 16% and 24%, respectively. These findings indicate a higher level of carbapenem resistance in *K. pneumoniae* than in *E. coli* and highlight the increasing concern of carbapenem-resistant Enterobacterales. (Dr. Gaurishanker Shrimali, et al., 2025)

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Conflicts of interest

There are no conflicts of interest.

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