

Detection of ESBL and Carbapenemase-Producing Enterobacteriaceae Using Phenotypic Methods

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Abstract:

Background: The rise of extended-spectrum beta-lactamase (ESBL) and carbapenemase-producing Enterobacteriaceae is a significant danger to world health, attributed to restricted treatment alternatives and heightened morbidity and mortality rates. Timely identification of these resistance mechanisms is crucial for effective antibiotic treatment and infection management.

Objectives: To identify ESBL and carbapenemase-producing Enterobacteriaceae by phenotypic approaches over the course of one year.

Methods: A laboratory-based cross-sectional investigation was performed over one year, encompassing 235 unique Enterobacteriaceae isolates sourced from diverse clinical cases. Isolates were detected with standard microbiological methods. Antimicrobial susceptibility testing was conducted using the Kirby–Bauer disc diffusion method. ESBL production was identified by the combined disc diffusion test (CDDT), whilst carbapenemase production was determined using the Modified Hodge Test (MHT) and the Carba NP test. The data were examined employing descriptive statistics.

Results: Of the 235 Enterobacteriaceae isolates, 92 (39.1%) were identified as producers of extended-spectrum beta-lactamases (ESBL), while 28 (11.9%) were identified as makers of carbapenemases. *Escherichia coli* was the primary bacteria creating extended-spectrum beta-lactamases, whereas *Klebsiella pneumoniae* was the most prevalent generator of carbapenemases. Significant resistance was noted against third-generation cephalosporins and fluoroquinolones, although colistin and tigecycline maintained effective action.

Conclusion: A considerable percentage of Enterobacteriaceae isolates were makers of ESBL and carbapenemase. Systematic phenotypic screening and rigorous antimicrobial stewardship are essential to mitigate the proliferation of multidrug-resistant pathogens.

Keywords: ESBL, Carbapenemase, Enterobacteriaceae, Phenotypic detection, Antimicrobial resistance.

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Introduction

Enterobacteriaceae are prevalent etiological agents of both community-acquired and nosocomial illnesses [1]. The extensive and frequently improper utilization of beta-lactam antibiotics has resulted in the development of beta-lactamase-mediated resistance, notably extended-spectrum beta-lactamases (ESBLs) and carbapenemases [2]. ESBL-producing organisms indicate resistance to penicillins, cephalosporins, and aztreonam, whereas carbapenemase-producing Enterobacteriaceae (CPE) demonstrate resistance to carbapenems, typically regarded as last-resort antibiotics [3].

India has documented a substantial prevalence of ESBL and carbapenemase manufacturers, greatly contributing to the worldwide antimicrobial resistance crisis [4]. Phenotypic detection technologies continue to be extensively utilized in regular diagnostic laboratories owing to their economic efficiency and practicality [5]. This study sought to ascertain the prevalence of ESBL and carbapenemase-producing Enterobacteriaceae by phenotypic approaches over the course of one year.

Materials and Methods

Study Design and Duration: This laboratory-based cross-sectional investigation was performed over one year at Bhagwan Mahavir Institute of Medical Sciences, Pawapuri, Nalanda.

Sample Size and Bacterial Isolates: A total of 235 unique Enterobacteriaceae isolates were obtained from clinical specimens, including urine, blood, pus, sputum, and bodily fluids.

Identification of Isolates: Isolates were identified by standard microbiological techniques, including colony morphology, Gram staining, and biochemical assays.

Antimicrobial Susceptibility Testing: Antimicrobial susceptibility testing was conducted using the Kirby–Bauer disc diffusion method on Mueller–Hinton agar and evaluated according to CLSI recommendations.

Phenotypic Detection of ESBL: ESBL generation was identified utilizing the combined disc diffusion test (CDDT) with ceftazidime and ceftazidime-clavulanic acid discs. A zone diameter increase of ≥ 5 mm in the presence of clavulanic acid was deemed positive.

Phenotypic Detection of Carbapenemase: Carbapenemase production was identified with the Modified Hodge Test (MHT) and the Carba NP test for isolates exhibiting diminished sensitivity to carbapenems.

Data Analysis: Data were inputted into Microsoft Excel and evaluated utilizing descriptive statistics. Results were presented as frequencies and percentages.

Results

Distribution of Enterobacteriaceae Isolates

Table 1: Distribution of Enterobacteriaceae isolates (n = 235)

Organism	Number	Percentage (%)
<i>Escherichia coli</i>	109	46.3
<i>Klebsiella pneumoniae</i>	75	31.9
<i>Enterobacter</i> spp.	26	11.0
<i>Citrobacter</i> spp.	16	6.8
<i>Proteus</i> spp.	12	5.1

ESBL and Carbapenemase Production

Table 2: Prevalence of ESBL and carbapenemase production

Resistance mechanism	Number	Percentage (%)
ESBL producers	94	40
Carbapenemase producers	27	11.5
Non-producers	116	49.6

Organism-wise Distribution of Resistance

Table 3: Organism-wise distribution of ESBL and carbapenemase producers

Organism	ESBL positive	Carbapenemase positive
<i>E. coli</i>	55	8
<i>K. pneumoniae</i>	32	14
Others	10	8

Antibiotic Resistance Pattern

Table 4: Antibiotic resistance profile of ESBL and carbapenemase producers

Antibiotic	Resistance (%)
Cefotaxime	88.7
Ceftazidime	85.9
Ciprofloxacin	71.5
Imipenem	13.7
Colistin	3.3

Discussion

The current analysis reveals a significant prevalence of ESBL-producing Enterobacteriaceae (39.1%), aligning with findings from prior Indian research. The appearance of carbapenemase producers (11.9%) is particularly alarming since it restricts therapeutic alternatives and heightens the

probability of treatment failure. *Escherichia coli* continued to be the primary producer of extended-spectrum beta-lactamases, although resistance to carbapenems was more prevalent in *Klebsiella pneumoniae* [6].

Phenotypic techniques, like CDDT and MHT, continue to serve as valuable instruments for routine

detection in resource-constrained environments [7]. The significant resistance to frequently utilized antibiotics highlights the critical necessity for antimicrobial stewardship, rigorous infection control measures, and regular surveillance [8].

Conclusion

A significant percentage of Enterobacteriaceae isolates were makers of ESBL and carbapenemase. Routine phenotypic screening for these resistance mechanisms should be integrated into microbiology laboratory protocols. Prudent antibiotic utilization and effective infection control measures are crucial to mitigate the dissemination of multidrug-resistant Enterobacteriaceae.

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