Iranian Journal of Microbiology
Volume 17 Number 3 (June 2025) 390-396
DOI: http://doi.org/10.18502/ijm.v17i3.18821

ORIGINAL ARTICLE

Evaluation of the in vitro efficacy of antimicrobials against Enterobacterales with multiple carbapenemase enzymes

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Received: January 2025**, Accepted:** April 2025

ABSTRACT

Background and Objectives: High-dose of carbapenems and combination therapies with new β-lactam/β-lactamase inhibitors and polymyxin B/tigecycline have been considered for treatment of carbapenem resistant Enterobacterales infection. The research was conducted to evaluate the in vitro potency of aminoglycosides, ceftazidime/avibactam/aztreonam and tigecycline against isolates of *Enterobacteriaceae* with multiple carbapenemase enzymes.

Materials and Methods: 42 genotypically confirmed carbapenem resistant Enterobacterales (twenty-nine NDM producers, nine NDM and OXA-48 producers, three NDM and VIM producers and one NDM combined with VIM and OXA 48 producer) were included. Minimum inhibitory concentration for carbapenems, aminoglycosides and tigecycline was determined by Vitek 2. Ceftazidime/avibactam/aztreonam synergy was observed by disk diffusion methodology.

Results: The in vitro efficacy of aminoglycosides was observed against *Escherichia coli (E. coli)* isolates with NDM and VIM genes. Low tigecycline susceptibility was observed among *Klebsiella pneumoniae (K. pneumoniae)* isolates with NDM and OXA-48 genes. Ceftazidime -avibactam/aztreonam combination displayed good in vitro activity against dual carbapenemase producers of *E. coli* isolates (NDM with OXA-48 and NDM with VIM genes) and *Klebsiella pneumoniae* (combination of NDM, VIM and OXA-48 genes).

Conclusion: Ceftazidime/avibactam/aztreonam, aminoglycosides and tigecycline displayed in vitro activity against dual carbapenemase producers of *E. coli* and *K. pneumoniae.*

Keywords: Enterobacterales; Dual carbapenemase; In vitro activity; Metallobetalactamases

INTRODUCTION

Carbapenems are frequently employed as initial treatments for severe infections involving gram negative bacteria, the wide spread occurrence of carbapenem resistant *Enterobacteriaceae* has restricted options for clinicians, forcing them to turn to non-beta lactam drugs as alternatives (1). While Metallo-Beta-Lactamases can break down all β-lactams except aztreonam, the later cannot be administered as monotherapy because of the co-production of expanded-spectrum-β-lactamases (ESBL). Presence of New Delhi metallo-β-lactamases (NDM) pose a notable challenge due to its broad hydrolytic activity

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on penicillins, cephalosporins, and carbapenem (2).

The term carbapenem-resistant *Enterobacteriaceae* (CRE) refers to isolates that demonstrates resistance to at least one of the carbapenem antibiotics or synthesize a carbapenemase. The number of Enterobacterial strains that produce multiple carbapenemases is steadily increasing (3).

The novel antibiotics currently approved for combating CRE infections comprise of imipenem/ cilastatin-relebactam, meropenem-vaborbactam, ceftazidime/avibactam (CZA), plazomicin and eravacycline (4). The currently leading therapeutic alternative for CRE are tigecycline, colistin, and ceftazidime/ avibactam (5). Ceftazidime/avibactam combination shows good bactericidal ability against KPC and OXA-enzyme producers when combined with aztreonam and enhances the synergistic antibacterial activity against NDM, IMP, KPC, IMP co-producers, and KPC combined with NDM producers (6).

For treatment of infections caused by CRE, high-dose and combination approaches including the new β-lactam/β-lactamase inhibitors should be considered (7). Against NDM and OXA dual producers, the bactericidal effect of polymyxin B/ tigecycline combination with β-lactams are variable in activity (8). This research was undertaken to find out the in vitro potency of antimicrobial agents against isolates with NDM, OXA-48 and VIM genes.

MATERIALS AND METHODS

A cross-sectional study was done on 42 carbapenem resistant isolates obtained from various clinical samples collected during October 2022 to January 2024. This study included twenty-nine NDM producers, nine NDM and OXA-48 co- producers, three NDM with VIM producers and one NDM, VIM and OXA-48 co-producers. This study was undertaken upon receiving Institutional Ethics Committee clearance (Ref No: VMKVMC and H/IEC/22/176 Dated 16/11/ 2022).

Kirby-Bauer disc diffusion method was employed for antimicrobial susceptibility testing using the following discs (Hi Media Laboratories Pvt. Ltd., Mumbai, India); amikacin (30 micrograms), amoxyclav (20/10 micrograms), aztreonam (30 micrograms), cefoperazone/sulbactam (75/30 micrograms), cefotaxime (30 micrograms), cefepime (30 micrograms), ceftazidime (30 micrograms), ceftazidime/avibactam (30 µg/20 micrograms), ciprofloxacin (5 micrograms), co-trimoxazole (25 micrograms), gentamicin (10 micrograms), imipenem (10 micrograms), meropenem (10 micrograms) and piperacillin/tazobactam (100/10 micrograms).

An inhibition zone of ≤ 19 mm for imipenem and meropenem was considered as resistant for *Enterobacteriaceae.* Ceftazidime/avibactam zone diameter of ≥ 21 mm and ≤ 20 mm was considered as susceptible and resistance, respectively. Aztreonam inhibition zone of ≥ 21 mm and ≤ 17 mm was considered as susceptible and resistance for Enterobacterales, respectively (9). Forty-two isolates which exhibited resistance to either imipenem, meropenem or both were characterized phenotypically by Vitek-2 automated system. Turbidometrically controlled suspension of organisms was used to inoculate Vitek 2 ID/AST Gram negative cards (AST-N405) and minimum inhibitory concentration of the antimicrobials were determined.

The confirmation of carbapenem resistant isolates at genotypic level was done by Hi-PCR Carbapenemase Gene (Multiplex) Probe PCR Kit (Himedia Laboratories, Mumbai). Carbapenemase genes (KPC, IMP, NDM, VIM, OXA-51, OXA-23, OXA-48 and OXA-58) were identified by Real Time PCR: Initial denaturation was carried out at 95°C for ten minutes under the following cycling protocol; Denaturation cycle at 95°C for five seconds and subsequent annealing and extension cycle at 60°C for one minute. The threshold cycle (Ct) value of less than or equal to 40 was regarded as positive for carbapenemase producers. Lack of amplification curve in the target genes channel was considered as negative.

Ceftazidime/avibactam/aztreonam synergy testing. Aztreonam (30 µg) disk was placed on ceftazidime/ avibactam (30 µg/20 µg) in culture suspension swabbed on Muller Hinton agar and incubated at 37°C overnight for 16-18 hours. Zone of inhibition diameter for ceftazidime/avibactam/aztreonam combination was analyzed as per CLSI guidelines (9). Statistical analysis was done by using frequencies and percentages.

RESULTS

Carbapenem resistant *E. coli* showed a minimum inhibitory concentration of 4 to ≥ 64 µg/ml for imipenem and meropenem. Carbapenem resistant *K. pneu-*

moniae showed a minimum inhibitory concentration of ≤ 2 to 16 µg/ml for imipenem and meropenem. Carbapenem resistant *Enterobacter* cloacae showed a minimum inhibitory concentration of 4 to 16 µg/ml for imipenem and meropenem, whereas *Proteus mirabilis* exhibited minimum inhibitory concentration of 16 µg/ml for carbapenems (Table 1).

Carbapenem resistant Enterobacterales showed resistance to ciprofloxacin (MIC: ≥ 4 microgram/ml). Amikacin resistant *E. coli* isolates exhibited a MIC of 32 to ≥ 64µg/ml, whereas *K. pneumoniae, E. cloacae* and *P. mirabilis* showed a MIC of ≥ 32 µg/ml. Gentamicin resistant Enterobacterales displayed a MIC of 8 to ≥ 16 µg/ml. Five (27.78%) isolates of *E. coli* with NDM gene and a single *E. coli* isolate with NDM and VIM gene showed susceptibility to amikacin (MIC:2 to 16 µg/ml). A single isolate of *K. pneumoniae* and *E. cloacae* with NDM gene showed susceptibility to amikacin (MIC: 2 to 16 µg/ml).

Five (27.78%) isolates of *E. coli* with NDM gene and a single *E. coli* isolate with NDM and VIM gene showed susceptibility to gentamicin (MIC: 1 to 2µg/ml). Four isolates each of *K. pneumoniae* with NDM and NDM+OXA-48 and a single isolate of *E. cloacae* with NDM gene showed susceptibility to gentamicin (MIC: 1 to 2 µg/ml) (Table 2).

17 (94.44%) out of 18 *E. coli* isolates with NDM gene showed resistance to ceftazidime/avibactam. *E. coli* with NDM plus OXA-48 gene and NDM plus VIM gene exhibited resistance to ceftazidime/avibactam. 2 (11.11%) out of the 18 *E. coli* isolates with NDM showed intermediate resistance to aztreonam. 16 (88.89%) out of the 18 *E. coli* isolates with NDM showed aztreonam resistance. *E. coli* with NDM and OXA-48 and NDM and VIM genes exhibited resistance to aztreonam. 12 (66.67%) *E. coli* with NDM gene showed synergism against ceftazidime/avibactam/aztreonam combination. *E. coli* with NDM plus OXA-48 and NDM genes plus VIM gene exhibited synergism against ceftazidime/avibactam/aztreonam combination (Table 3).

A single isolate of *K. pneumoniae* with NDM alone & NDM plus VIM gene showed susceptibility to ceftazidime/avibactam. Two isolates of *K. pneumoniae* with NDM plus OXA-48 showed susceptibility to ceftazidime/avibactam. *K. pneumoniae* isolates with NDM plus OXA -48 exhibited resistance to aztreonam. A single isolate of *K. pneumoniae* with NDM plus VIM gene showed intermediate resistance to aztreonam. Six out of the seven *K. pneumoniae* isolates with NDM gene exhibited synergism against ceftazi-

Table 1. Minimum inhibitory cncentration of carbapenems against Enterobacterales

Organisms	**Minimum inhibitory concentration**						
	Imipenem				**Meropenem**		
No of isolates (n=42)	≥64 µg/ml (R)	≥16 µg/ml (R)	4-8 µg/ml (R)	≤2 µg/ml (I)	≥64 µg/ml (R)	≥16 µg/ml (R)	4-8 µg/ml (R)
NDM *E. coli* (n=18)	1 (5.56%)	9 (50.00%)	8 (44.44%)	0 (0%)	0 (0%)	14 (77.78%)	4 (22.22%)
E. coli (n=3) (Co-producer of NDM & OXA -48)	0 (0%)	1 (33.33%)	2 (66.67%)	0 (0%)	0 (0%)	1 (33.33%)	2 (66.67%)
E. coli (n=2) (Co-producer of NDM & VIM)	0 (0%)	1 (50.00%)	1 (50.00%)	0 (0%)	0 (0%)	2 (100%)	0 (0%)
NDM *K. pneumoniae* (n=7)	0 (0%)	5 (71.43%)	2 (28.57%)	0 (0%)	0 (0%)	5 (71.43%)	2 (28.57%)
K. pneumoniae (n=5) (Co-producer of NDM & OXA-48)	0 (0%)	1 (20.00%)	3 (60.00%)	1 (20.00%)	0 (0%)	5 (100%)	0 (0%)
K. pneumoniae (n=1) (Co-producer of NDM& VIM)	0 (0%)	0 (0%)	1 (100%)	0 (0%)	0 (0%)	1 (100%)	0 (0%)
K. pneumoniae (n=1) (Co-producer of NDM, VIM & OXA-48)	0 (0%)	1 (100%)	0 (0%)	0 (0%)	0 (0%)	1 (100%)	0 (0%)
E. cloacae (n=4) (NDM)	0 (0%)	1 (25.00%)	3 (75.00%)	0 (0%)	0 (0%)	2 (50.00%)	2 (50.00%)
P. mirabilis (n=1) (NDM & OXA-48)	0 (0%)	1 (100%)	0 (0%)	0 (0%)	0 (0%)	1 (100%)	0 (0%)

*R-Resistant

*I-Intermediately Resistant

Table 2. Minimum inhibitory concentration of aminoglycosides against carbapenem resistant Enterobacterales

Organisms	Minimum inhibitory concentration					
	Amikacin			Gentamicin		
No of isolates (n=42)	≥64 µg/ml (R)	≥32 µg/ml (I)	2-16 µg/ml (S)	≥16 µg/ml (R)	8 µg/ml (I)	1-2 µg/ml (S)
E. coli (n=18) (NDM)	13 (72.22%)	0 (0%)	5 (27.78%)	13 (72.22%)	0 (0%)	5 (27.78%)
E. coli (n=3) (NDM& OXA -48)	2 (66.67%)	1 (33.33%)	0 (0%)	2 (66.67%)	1 (33.33%)	0 (0%)
E. coli (n=2) (NDM & VIM)	1 (50%)	0 (0%)	1 (50%)	1 (50%)	0 (0%)	1 (50%)
K. pneumoniae (n=7) (NDM)	0 (0%)	6 (85.71%)	1 (14.29%)	3 (42.86%)	0 (0%)	4 (57.14%)
K. pneumoniae (n=5) (NDM & OXA-48)	0 (0%)	5 (100%)	0 (0%)	1 (20.00%)	0 (0%)	4 (80.00%)
K. pneumoniae (n=1) (NDM& VIM)	0 (0%)	1 (100%)	0 (0%)	1 (100%)	0 (0%)	0 (0%)
K. pneumoniae (n=1) (NDM,VIM & OXA-48)	0 (0%)	1 (100%)	0 (0%)	1 (100%)	0 (0%)	0 (0%)
E. cloacae (n=4) (NDM)	0 (0%)	3 (75.00%)	1 (25.00%)	3 (75.00%)	0 (0%)	1 (25.00%)
P. mirabilis (n=1) (NDM & OXA-48)	0 (0%)	1 (100%)	0 (0%)	1 (100%)	0 (0%)	0 (0%)

Table 3. In vitro activity of ceftazidime/avibactam/aztreonam combination against carbapenem resistant *E. coli*

S. No	Sample ID	Zone diameter in millimeter			Interpretation	Genes detected
		Ceftazidime/Avibactam	Aztreonam	Ceftazidime/ Avibactam/ Aztreonam	Synergy/No synergy	
1	Pus	R (9 mm)	I (19 mm)	19 mm	Synergy negative	NDM
2	Pus	R (6 mm)	R (6 mm)	6 mm	Synergy negative	NDM
3	Urine	S (24 mm)	R (6 mm)	19 mm	Synergy negative	NDM
4	Urine	R (6 mm)	R (6 mm)	6 mm	Synergy negative	NDM
5	Urine	R (14 mm)	R (16 mm)	19 mm	Synergy positive	NDM&OXA 48
6	Urine	R (20 mm)	R (6 mm)	20 mm	Synergy positive	NDM
7	Urine	R (14 mm)	R (6 mm)	20 mm	Synergy positive	NDM&OXA 48
8	Pus	R (6 mm)	R (11 mm)	23 mm	Synergy positive	NDM
9	Pus	R (14 mm)	R (6 mm)	17 mm	Synergy positive	NDM
10	Urine	R (6 mm)	R (6 mm)	16 mm	Synergy positive	NDM
11	Urine	R (16 mm)	R (6 mm)	20 mm	Synergy positive	NDM
12	Pus	R (14 mm)	R (6 mm)	17 mm	Synergy positive	NDM &VIM
13	Pus	R (12 mm)	R (10 mm)	19 mm	Synergy positive	NDM
14	Urine	R (16 mm)	R (6 mm)	21 mm	Synergy positive	NDM&OXA 48
15	Urine	R (12 mm)	R (13 mm)	22 mm	Synergy positive	NDM
16	Urine	R (12 mm)	R (16 mm)	12 mm	Synergy negative	NDM
17	Urine	R (11 mm)	R (12 mm)	13 mm	Synergy positive	NDM
18	Urine	R (14 mm)	R (6 mm)	18 mm	Synergy positive	NDM&VIM
19	Urine	R (14 mm)	R (6 mm)	14 mm	Synergy positive	NDM
20	Urine	R (14 mm)	R (6 mm)	17 mm	Synergy positive	NDM
21	Urine	R (15 mm)	R (6 mm)	15 mm	Synergy positive	NDM
22	Urine	R (15 mm)	I (18 mm)	10 mm	Synergy negative	NDM
23	Urine	R (10 mm)	R (10 mm)	15 mm	Synergy positive	NDM

dime/avibactam/aztreonam combination. Three out of the five *K. pneumoniae* isolates with NDM plus OXA-48 gene exhibited synergism and a single isolate of *K. pneumoniae* with NDM plus VIM gene plus OXA-48 exhibited synergism against ceftazidime/avibactam/aztreonam combination (Table 4).

Isolates of *E. cloacae* with NDM gene showed resistance to ceftazidime/avibactam alone and aztreo-

nam and exhibited ceftazidime/avibactam/aztreonam synergism. A single isolate of *P. mirabilis* with NDM plus OXA-48 showed ceftazidime/avibactam resistance and intermediate resistance to aztreonam but exhibited ceftazidime/avibactam/aztreonam combination synergism (Table 5).

A single isolate of *E. cloacae* and *K. pneumoniae* with NDM, and a single isolate of *E. coli* with NDM plus OXA-48 showed susceptibility to cotrimoxazole (MIC:≤20 µg/ml). Three isolates of *K. pneumoniae* with NDM showed resistance to tigecycline (4 to ≥8 µg/ml). A single isolate of *E. coli* with NDM showed tigecycline resistance with MIC of 4 µg/ml. Five isolates of *K. pneumoniae* with NDM plus OXA-48 showed resistance to tigecycline (2 to ≥ 8µg/ml). A single isolate of *E. cloacae* with NDM showed tigecycline resistance with MIC of 2 µg/ml.

DISCUSSION

The efficacy of antimicrobial agents varies against carbapenem resistant isolates due to its dual carbapenemase enzymes. Our study revealed that 30.95% of isolates carried multiple enzymes (NDM plus OXA-48 genes, NDM plus VIM genes, NDM, VIM plus OXA-48 genes). No KPC genes were detected among the isolates during the study period. Literature has shown co-production of NDM and OXA-48 genes in 27% of carbapenem resistant clinical isolates (10).

In our study, carbapenem resistant *K. pneumoniae* isolates showed a MIC of ≥ 16 µg/ml for meropenem and imipenem. These isolates carry either NDM plus VIM plus OXA-48 genes or NDM plus OXA-48 genes. Research has shown that carbapenem resistant

Table 4. In vitro activity of ceftazidime/avibactam/aztreonam combination against carbapenem resistant *K. pneumoniae*

S. No	Sample ID	Zone diameter in millimeter			Interpretation Synergy/No synergy	Genes Detected
		Ceftazidime/ Avibactam	Aztreonam	Ceftazidime/ Avibactam/ Aztreonam		
1	Urine	S (25 mm)	I (20 mm)	28 mm	Synergy negative	NDM & VIM
2	Endotracheal aspirate	S (25 mm)	R (6 mm)	26 mm	Synergy negative	NDM & OXA 48
3	Pus	R (12 mm)	R (6 mm)	19 mm	Synergy positive	NDM
4	Endotracheal aspirate	R (6 mm)	R (6 mm)	29 mm	Synergy positive	NDM & OXA 48
5	Endotracheal aspirate	R (6 mm)	R (6 mm)	29 mm	Synergy positive	NDM & OXA 48
6	Pus	R (6 mm)	R (17 mm)	29 mm	Synergy positive	NDM
7	Pus	S (25 mm)	R (6 mm)	28 mm	Synergy negative	NDM
8	Urine	R (12 mm)	R (6 mm)	22 mm	Synergy positive	NDM
9	Endotracheal aspirate	R (10 mm)	R (12 mm)	14 mm	Synergy positive	NDM & OXA 48
10	Pus	R (12 mm)	R (6 mm)	22 mm	Synergy positive	NDM
11	Urine	S (24 mm)	R (6 mm)	22 mm	Synergy negative	NDM & OXA 48
12	Urine	R (12 mm)	R (6 mm)	25 mm	Synergy positive	NDM, VIM & OXA 48
13	Urine	R (12 mm)	R (11 mm)	18 mm	Synergy positive	NDM
14	Urine	R (12 mm)	R (12 mm)	25 mm	Synergy positive	NDM

Table 5. In vitro activity of ceftazidime/avibactam/aztreonam combination against carbapenem resistant *E. cloacae* and *P. mirabilis*

S. No	Sample ID	Organisms	Zone diameter in millimeter			Interpretation Synergy/ No synergy	Genes Detected
			Ceftazidime/ Avibactam	Aztreonam	Ceftazidime/ Avibactam/ Aztreonam		
1	Pus	*Enterobacter cloacae*	R (15 mm)	R (16 mm)	22 mm	Synergy positive	NDM
2	Pus	*Proteus mirabilis*	R (14 mm)	I (19 mm)	25mm	Synergy positive	NDM OXA 48
3	Urine	*Enterobacter cloacae*	R (14 mm)	R (11 mm)	27 mm	Synergy positive	NDM
4	Urine	*Enterobacter cloacae*	R (6 mm)	R (9 mm)	14 mm	Synergy positive	NDM
5	Urine	*Enterobacter cloacae*	R (10 mm)	R (6 mm)	19 mm	Synergy positive	NDM

(meropenem MIC > 16 µg/ml) *K. pneumoniae* isolates carry NDM plus OXA-48 genes (11).

In the present study, ceftazidime/avibactam/aztreonam combination synergy was observed against 100 % of the *E. coli* isolates with multiple carbapenemase enzymes (NDM plus OXA-48 genes & NDM plus VIM genes), 60 % *K. pneumoniae* isolates with NDM plus OXA-48 genes and a single isolate of *K. pneumoniae* (NDM plus OXA-48 plus VIM genes). A single *P. mirabilis* with NDM with OXA-48 showed ceftazidime/avibactam/aztreonam combination synergism. In the present study, MBL-producing strains were predominantly detected among clinical isolates and ceftazidime/avibactam/aztreonam combination showed synergism against NDM,VIM & OXA-48 isolates. The present study did not evaluate the in vivo efficacy of ceftazidime/avibactam/aztreonam combination. The treatment of MBL producing strains by ceftazidime/avibactam/aztreonam have shown success in several clinical cases (12). The combination of ceftazidime/avibactam/aztreonam showed remarkable effectiveness against MBL-producing *K. pneumoniae* including co-producers of more than one carbapenemase in an in vitro study (13).

In the present study amikacin and gentamicin susceptibility was observed among 50% of the *E. coli* isolates which were co-producers of NDM plus VIM. 100% of the *K. pneumoniae* isolates with multiple carbapenemase enzymes showed intermediate resistance to amikacin (≥ 32 µg/ml) and 80% of the *K. pneumoniae* (NDM and OXA-48) showed gentamicin susceptibility (1 to 2 µg/ml). 76.19% of the isolates in our study showed tigecycline susceptibility (≤ 0.5 to 1 µg/ml).

Studies have demonstrated the in vitro efficacy of ceftazidime/avibactam and imipenem, meropenem, amikacin and tigecycline combination. An in vitro study on the antimicrobial efficacy of ceftazidime/avibactam when combined with ertapenem, imipenem, meropenem, gentamicin, tigecycline, and ciprofloxacin against carbapenemase producer *K. pneumoniae* showed that ceftazidime/avibactam and imipenem or meropenem combination had synergistic activity (14). In a study on carbapenem resistant *K. pneumoniae*, the combination of meropenem and ceftazidime/avibactam against NDM-producing strains showed synergy rate of 57.14% and 91.67% against IMP-producing strains (15). The combination of ceftazidime/avibactam and meropenem seems to have a synergistic effect against multidrug-resistant *K. pneumoniae* (16).

When combined with amikacin and tigecycline, ceftazidime/avibactam demonstrated enhanced antibacterial effects compared to single drug therapy (17). By disrupting membrane permeability and protein synthesis of bacteria, aminoglycosides can enhance the bactericidal effects of ceftazidime/avibactam and amikacin combination leading to resistance prevention. Ceftazidime/avibactam plus gentamicin combination was found to be synergistic against 27% of the isolates in a study, whereas combinations with tigecycline resulted in no synergistic killing against any of the isolates and demonstrated antagonism against 13% of the isolates (18).

The in vitro efficacy of ceftazidime/avibactam/aztreonam, aminoglycosides and tigecycline against dual carbapenemase producers of *E. coli* and *K. pneumoniae* was observed in our study. Ceftazidime/avibactam/aztreonam combination displayed good in vitro activity against dual carbapenemase producers of *E. coli* isolates (NDM with OXA-48 co-producers and NDM and VIM co-producers) and *K. pneumoniae* (NDM,VIM plus OXA-48). *K. pneumoniae* isolates with NDM plus OXA-48 genes showed low susceptibility to tigecycline. In our research, the in vitro efficacy of aminoglycosides was observed against *E. coli* isolates with NDM and VIM genes.

CONCLUSION

Predicting the in vivo efficacy based on the in vitro susceptibility of amikacin, gentamicin, ceftazidime/avibactam and tigecycline is not always feasible because of factors like pharmacodynamics and host immune response that can influence the clinical outcome of the patient. The genomic analysis of the study detected only VIM, NDM and OXA-48 in Enterobacterales and hence the in vitro susceptibility of antimicrobials was tested only among strains carrying VIM, NDM and OXA-48 genes.

Limitation of the study. Presence of multiple carbapenemase was observed only among 30.95% of the isolates included in the study. Detection of in vitro activity of antimicrobials against carbapenem resistant genes other than VIM, NDM and OXA-48 was beyond the scope of our study.

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