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Analysis of rectal carbapenem-resistant *Enterobactericeae* colonization results first report in Istanbul/Turkiye: Klebsiella pneumoniae co-producing $bla_{KPC} + bla_{NDM} + bla_{OXA-48}$ in a single strain

Kamuran Şanlı[1*] and Beyza Öncel[1]

Abstract

Background Carbapenemase-Producing *Enterobacteriaceae* (CPE) are known to be on the rise globally and are a major concern. Rapid screening of at-risk patients and implementation of infection control measures can prevent colonization from becoming a source of infection.

Objective We investigated the incidence of carbapenem-resistant *Enterobacteriaceae* (CRE) colonization, CRE-producing *Enterobacteriaceae* species, carbapenemase gene types and prevalence in patients subjected to CRE screening tests in rectal swab samples using the CHROMAgar culture method and real-time Polymerase Chain Reaction (PCR) method in a tertiary hospital in Istanbul, Turkiye. In addition, we investigated the effectiveness of diagnosis with PCR and CHROMAgar methods by examining whether meropenem-resistant infection foci developed in patients with CRE colonization from hospitalization to discharge.

Materials and methods This study was conducted retrospectively in the Microbiology Laboratory of 3953 patients at Başakşehir Çam and Sakura City Hospital in Istanbul between January 2021 and December 2023. All data, including CRE colonization screening test results using CHROMAgar and PCR methods, age, gender, requesting clinic, and meropenem-resistant *Enterobacteriaceae* culture results in infection foci that developed during hospitalization, were obtained from the hospital automated system.

Results In all 3953 patients included in the study, CRE colonization was screened by CHROMAgar culture method on rectal swab samples and by real-time PCR method in 500 patients. The mean age of the patients was 42.9 ± 30.1 and 58.1% were male. There was no difference in age and gender distribution between the Upon admission to the hospital and during hospitalization groups ($p > 0.05$). Rectal CRE colonization was detected in 9.7% of all patients with the CROMAgar method and in 9.2% with the PCR method and in 16.2%. The most statistically significant increase was seen in intensive care units with the CHROMAgar method ($p < 0.05$). Among *Enterobacteriaceae* species, *Klebsiella pneumoniae* (69.6%) was most frequently grown, followed by *Escherichia coli* (13.7%). There was no statistically significant difference between the groups in terms of CRE growth status and microorganism distribution in rectal swab samples of patients during hospitalization and hospital admission ($p > 0.05$). Among the CRE genes detected

*Correspondence:
Kamuran Şanlı
kamuran67@gmail.com

by PCR, single genes were the most common. While OXA-48 positivity was detected in 113 (16.40%) of the patients, bla_{NDM} was detected in 31 (4.50%), bla_{KPC} was detected in 5 (0.73%) and $bla_{VIM/IMP}$ gene was detected in 5 (0.73%) of the others. Double gene positivity was observed in a single strain in 6 patients and bla_{KPC}, bla_{NDM} and bla_{OXA-48} genes were positive together in a single strain in 3 patients. Infection developed in one or more foci resistant to carbapenem in 159 of the patients colonized with CRE during their hospitalization. The rate of infection development was lower in patients colonized by PCR compared to chromogenic agar.

Conclusion The CRE colonization rate is high and should be closely monitored with infection control precautions. Although the detection rate of CRE colonization was higher with PCR, the rate of progression to infection in any focus during hospitalization was found to be lower detected with the CHROMAgar method. This situation showed that rapid detection of CRE colonization with PCR, early diagnosis and isolation are important. *Klebsiella pneumoniae* colonization was the most common causative agent. Co-production of 3 CRE genes ($bla_{KPC} + bla_{NDM} + bla_{OXA-48}$) in 3 patients in our hospital highlighted the importance of concerns about multi-resistant Enterobacteriaceae.

Keywords Carbapenem-resistant *Enterobacteriaceae*, *Klebsiella pneumoniae*, Polymerase chain reaction, *Enterobacteriaceae*, Hospitalization

Introduction

Enterobacteriaceae are Gram-negative bacteria that are predominantly detected in soil, water, the intestines of humans and many animals. Although they are a component of the flora, they can cause bacteremia, urinary tract infections (up to 70%), and intestinal infections [1]. Carbapenems are widely acknowledged for their efficacy in the management of multidrug-resistant infections. However, there is a mention in some studies of carbapenem resistance in infections caused by these bacteria [2]. *Enterobacteriaceae* hydrolyze carbapenem antibiotics via their carbapenemases, a β-lactamase group. Three main classes of carbapenemases are prevalent in clinical CRE isolates globally. These include class A (mostly KPC), class B (VIM, NDM and IMP) and class D (OXA-48 and its variants, OXA-162 and OXA-181 etc.) [3, 4]. Carbapenemase genes in particular are carried by large conjugative plasmids, thus facilitating horizontal transfer of carbapenem resistance among different bacterial strains and species. OXA-48 type carbapenemases have been identified mostly from North African countries, the Middle East, Turkiye and India, which constitute the most important reservoirs. KPC is the most common carbapenemase produced by *Enterobacteriaceae* [5]. It has also been reported to be the most common carbapenemase in the United States in a domestic study. In addition, outbreaks of NDM-producing *Enterobacteriaceae* have been reported [6, 7].

The global dissemination of CPE poses a significant concern for healthcare services worldwide [8–10]. Therapeutic options for CPE are limited and these bacteria demonstrate a strong capability to spread these resistance mechanisms [11]. Various measures are taken to prevent transmission of CPE in hospitals. These include the adoption of hospital screening and surveillance protocols alongside stringent infection control measures. In order to prevent the spread of these bacteria in hospitals, screening tests for rectal CPE carriage are performed [12, 13] and colonized patients are actively monitored and isolated [14, 15]. Rectal CPE screening test methods such as the ertapenem disk method (on direct culture), ertapenem-containing selective broth enrichment, and CHROMAgar™ KPC have been used and recommended for this purpose. However, except for CHROMAgar™ KPC, isolation times can reach 96 h and delays detection and treatment [16–18]. Rapid demonstration of carbapenemase gene mutations in rectal swab samples via PCR has been confirmed to have reasonable power to accurately detect CPE carriers [19]. In recent years, CRE infections have increased in patients with comorbidities and in intensive care units. These cases are often severe and result in high mortality and morbidity rates, prolonged hospitalization, and higher management costs [2]. The prevalence of rectal colonization with CPE among patients has been previously reported, with rates ranging from 0.5% to 32.9% in various regions worldwide [20–25].

Data on the prevalence of CPE colonization from Türkiye are limited. No previous study has been conducted on rectal CRE colonization in our hospital. This study was designed as the first study. We aimed to analyze the prevalence of CPE colonization, common *Enterobacteriaceae* species, and the distribution of detected genes in rectal swab samples obtained from patients during and after hospitalization at a tertiary healthcare institution in Istanbul by PCR and CHROMAgar methods. We also investigated the prevalence of developing a focus of infection due to carbapenem-resistant *Enterobacteriaceae* bacteria in patients with any CRE colonization during hospitalization and compared the rates of infection development among patients diagnosed with rectal

colonization by CHROMAgar and PCR. We observed the effect of early diagnosis of rectal carriage on the rate of infection development.

Material and methods

Data sources

This study was conducted as a retrospective study in the Department of Medical Microbiology at the 2648-bed Başakşehir Çam Sakura City Hospital. All patients who were hospitalized and treated as inpatients between from January 2021 to December 2023 and who were asked for CRE screening in rectal swab samples by culture and PCR method were included in the study. The ages and genders of the patients included in the study, the clinic from which the tests were requested, whether the tests were requested during outpatient clinic application or during inpatient treatment, the CRE results in the rectal swab culture test requested for each patient, and the carbapenem resistance gene results obtained by PCR in the rectal swab sample were obtained from the hospital automation system. In addition, bacterial growth and antimicrobial results were recorded in any culture material such as urine, wound, blood, sterile body fluid, and tracheal aspirate taken during hospitalization of all patients to screen whether CRE-positive patients developed a carbapenem-resistant infection focus during their hospitalization.

Upon admission to the hospital

It defines the patient group that undergoes rectal CRE colonization testing in patients who are referred from another hospital and admitted to our hospital or in patients who have not yet been admitted to our hospital but are planned to be admitted to the hospital.

During hospitalization

It defines the group of patients who are hospitalized in our hospital and have been tested for CRE colonization at any time.

Our hospital's rectal CRE colonization test policy

In our hospital; CRE screening test using the CHROMAgar culture method on rectal swabs is performed in all patients. The PCR method is only used in more critical patient groups such as liver transplant units. Because, ESCMID; "According to local epidemiology, we recommend rectal screening to identify CRE carriers before liver transplant surgery" and "According to local epidemiology, it would be good clinical practice to screen all solid organ transplant recipients for CRE before surgery" [26] definitions were used by liver transplant clinics and received administrative permission.

In addition, contact precautions are taken for rectal CRE carriers in our hospital. For this; patient isolation, case communication, more care in nursing care, comprehensive environmental cleaning etc. are implemented.

CRE colonization test study using culture method in rectal swab sample

Rectal swab samples taken from all patients are inoculated on chromogenic agar plates (Chromogenic KPC Agar, RTA, Turkey) using the direct plating method in our microbiology laboratory, incubated in a 37 °C incubator for 18–24 h. Identification of Gram-negative organisms grown on selective medium is performed using Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS, EXS2600, Zybio, China). For Enterobacteriaceae group bacteria, isolates are prepared again at 0.5 McFarland standard turbidity and tested for carbapenem resistance using the disk diffusion method with 10 µg disks on Mueller–Hinton agar. Zone diameter measurements are evaluated in accordance with EUCAST criteria and carbapenem resistance is defined. (EUCAST recommends a threshold zone diameter of < 28 mm if meropenem is to be used as a screening test for carbapenemase production) CDC define meropenem as "the most balanced in terms of sensitivity and specificity" among all Carbapenemase in the CRE screening test. For this reason, meropenem is preferred in CRE screening tests in our clinic [27].

CRE colonization test study using PCR method in rectal swab sample

Rectal swab samples sent to our laboratory were studied using the real-time PCR method (BD MAX Check-Points CPO test company USA) Carbapenem resistance genes bla_{KPC}, bla_{NDM}, bla_{VIM}/bla_{IMP} and $bla_{\text{OXA-48}}$ were determined. The study protocol was carried out according to the manufacturer's recommendations and is summarized as follows: Rectal swab samples taken with dry swabs were homogenized and transferred to a buffer tube. The buffer tube was quickly transferred and placed in the instrument, which performed automatic bacterial cell lysis, DNA extraction and concentration, reagent dehydration, nucleic acid amplification and real-time PCR to detect the target nucleic acid sequence. Amplification targets were detected using hydrolysis probes labeled with quenched fluorophores. Positive and negative controls underwent the same procedures for each test. Signal amplification, detection and interpretation were performed automatically by the instrument.

Investigation of infection foci developing during hospitalization in patients with rectal colonization

In patients with positive CRE colonization in rectal swab samples, it was investigated whether a focus of infection developed at any time from hospitalization to discharge.

The fact that the bacteria grown in the patient's focus of infection were the same as those grown in his own rectal swab sample and that meropenem resistance was detected in his antimicrobial susceptibility profile suggested that the focus of infection may have originated from his own rectal microbiota. Therefore, all culture samples sent to our microbiology laboratory from these patients, such as urine, wound, blood, sterile body fluids and tracheal aspirate, and the antimicrobial susceptibility profiles of those with growth were examined.

Those sensitive to meropenem were not included in the study. In addition, Gram-negative and Gram-positive bacteria grown outside the Enterobacteriaceae family and their antimicrobial results were not taken into account. Repeated tests for the same patient were excluded.

Cultures requested from any focus; Each culture request was cultured according to the recommendations of classical microbiology guidelines. Identification of microorganisms growing to the species level in each culture was performed using the MALDI-TOF MS Antibiotic susceptibility testing was performed with a Phoenix M50 (Bruker Daltonics, USA) instrument with a Gram-negative panel for each Enterobactericea family bacteria identified.

Statistical analysis

The data of the study were analyzed using SPSS Version 26.0 statistical package program (*IBM Corp. Released 2019. IBM SPSS Statistics for Windows, Version 26.0. Armonk, NY: IBM Corp.).* Categorical variables were presented as numbers and percentages; continuous variables were presented as "mean ± standard deviation" and "median (minimum–maximum)" values. In comparisons between groups, Fisher's Exact Test and Chi-Square Test were used for categorical variables. Kolmogorov Smirnov test was used as the normality test, and since the continuous variable was not distribued normally, Mann–Whitney u test was used for continuous variables. The statistical significance level was accepted as "$p < 0.05$".

Results

Our study is a retrospective study. A total of 3953 patients who were screened for CRE colonization in rectal swab samples were included in the study. The mean age of the patients was 42.9 ± 30.1, the median was 51 (minimum 0-maximum 108), and 58.1% were male. There was no difference in age and gender distribution between screening tests performed during hospital admission and during hospitalization ($p > 0.05$) (Table 1).

All 3953 patients were screened for CRE colonization using the CHROMAgar culture method, and only 500 were screened for CRE colonization using the PCR method. CRE colonization positivity using the CHROMAgar method was higher in intensive care clinics (ICU) at a rate of 10%, and lower in other clinics at a rate of 6.7%. This rate was statistically significant ($p < 0.05$). Among intensive care patients; CRE positivity rate in pediatric ICU patients was higher than in other ICU units ($p < 0.05$). Among clinics outside the intensive care unit, CRE positivity rate was higher in patients hospitalized in the gastroenterology clinic (33.3%) and statistically significant ($p < 0.05$) (Table 2).

CRE colonization test screening with PCR test method was performed only in the liver transplantation clinic in our hospital, as it is a test allowed to be performed only in these clinics.

CRE screening was performed in rectal swab culture with CHROMAgar in 3953 of all patients. 3588 (90.8%) of these patients were CRE negative, and 365 patients (9.2%) were CRE positive. The most commonly detected microorganism was Klebsiella pneumoniae (69.6% of CRE positive patients), and the second was Escherichia coli (13.7%). There was no statistically significant difference

Table 1 Basic descriptive characteristics of patients

	All patients ($n = 3953$)	Upon admission to the hospital ($n = 66$) (1.7%)	During hospitalization ($n = 3887$) (98.3%)	p
Age				
Mean ± SD	42,9 ± 30,1	38,3 ± 17,6	42,9 ± 30,3	0,063[b]
Median (min–max)	51,0 (0–108)	35,5 (2–75)	52,0 (0–108)	
Sex *n* (%)[a]				
Male	2297 (58,1)	40 (60,6)	2257 (58,1)	0,708[b]
Female	1656 (41,9)	26 (39,4)	1630 (41,9)	

[a] Colon percentage

[b] Mann-Whitney U test (Upon admission to the hospital and During hospitalization were compared)

Table 2 CRE positivity according to general characteristics of the patients according CHROMAgar metod and PCR metod

	CROMAgar culture test			PCR test	
	All patients (*n* = 3953)	**Upon admission to the hospital (*n* = 66)**	**During hospitalization (*n* = 3887)**	**Upon admission to the hospital (*n* = 65)**	**During hospitalization (*n* = 435)**
	***N / n* (%)[a]**	***N / n* (%)[a]**	***N / n* (%)[a]**	***N / n* (%)[a]**	***N / n* (%)[a]**
Sex					
Male	2297/ 199 (8.7)	40 / 0	2257 /199 (8.8)	38 / 5 (13.1)	238 /44 (18.48)
Female	1656 / 166 (10.0)	26 /3 (11.5)	1630 /163 (10.0)	27 / 3 (11.1)	197 /29 (14.7)
	$p = 0.148$[b]	$p = 0.057$[c]	$p = 0.116$[c]	$p = 0.999$[c]	$p = 0.299$[b]
Clinic					
Intensive care	3030/303 (10.0)				
General intensive care	1840 / 202 (11.0)	-	1840 / 202 (11.0)	1 / 1 (100.0)	-
Pediatric intensive care	382 / 26 (6.8)	-	382 / 26 (6.8)	2 / 0	-
CVS intensive care	260 / 8 (3.1)	-	260 / 8 (3.1)	-	-
Pandemic intensive care	104/ 5 (4.8)	-	104/ 5 (4.8)	-	-
Pediatric cardiac intensive care	313 / 58 (18.5)	-	313 / 58 (18.5)	-	-
Burn intensive care unit	100 / 1 (1.0)	-	100 / 1 (1.0)	-	-
Liver transplant intensive care unit	14 / 0	-	14 / 0	-	15 / 0
Neurology intensive care	14/ 1 (7.1)	-	14/ 1 (7.1)	-	-
Palliative care	3 / 2 (66.7)	-	3 / 2 (66.7)	-	-
	$p < 0.0001$[b]	-	$p < 0.0001$[b]	p[d]	p[d]
Other	923/62 (6.7)				
Liver, bile duct, pancreas surgery clinic	352 / 25 (7.1)	64 / 3 (4.7)	288 / 22 (7.6)	55 / 4 (7.2)	298 / 54 (18.2)
Pediatric cardiology	213 / 15 (7.0)	-	213 / 15 (7.0)	-	-
Pediatric cardiovascular surgery	171 / 9 (5.3)	-	171 / 9 (5.3)	-	-
Liver transplant	111 / 2 (1.8)	-	111 / 2 (1.8)	-	112 / 17 (15.1)
Pediatric bone marrow transplant	27 / 2 (7.4)	-	27 / 2 (7.4)	1 / 0	-
General surgery	10 / 2 (20.0)	1 / 0	9 / 2 (22.2)	1 / 0	-
Gastroenterology	6 / 2 (33.3)	-	6 / 2 (33.3)	5 /3 (60.0)	-
Burn unit	6 / 0	-	6 / 0	-	-
Pediatric gastroenterology	5 / 0	-	5 / 0	-	-
Neurosurgery	5 / 1 (20.0)	-	5 / 1 (20.0)	-	-
Internal medicine	4 / 2 (50.0)	-	4 / 2 (50.0)	-	1 /1 (100)
Pediatric diseases	2 / 0	-	2 / 0	-	-
Child emergency	2 / 0	1/ 0	1 / 0	-	-
Other	9 / 2 (22.2)	-	9 / 2 (22.2)		9 / 1 (11.1)
	$p = 0.002$[b]	$p = 0.952$[c]	$p = 0.002$[b]	p[d]	p[d]

[a] The percentage of CRE positive is given

[b] Chi-square test

[c] Fisher's exact test

[d] Incalculable

between the groups in terms of CRE growth and microorganism distribution in the CHROMAgar test of Upon admission to the hospital and durring hospitalization ($p > 0.05$) (Table 3).

Of all patients, only 500 had CRE screening test by PCR in rectal swab samples. CRE colonization positivity was detected in 81 of these patients (16.2%). These 81 patients, the "bla_{KPC}" gene 6.1%, the "bla_{NDM}" gene 21.0%; the "bla_{OXA-48}" gene 84.0%, and the "bla_{VIM}/bla_{IMP}" gene 2.5%; was positive. More than one gene was detected in nine bacterial isolates. These were; In one patient, the "bla_{KPC}" and "bla_{OXA-48}" genes

Table 3 Rectal swab culture and PCR screening test results

	All patients (*n* = 3953) *n*(%)[a]	Upon admission to the hospital (*n* = 66) *n*(%)[a]	During hospitalization (*n* = 3887) *n*(%)[a]	p
CRE results with CHROMAgar				
Negative	3588 (90.8)	63 (95.5)	3525 (81.7)	0,128[c]
Positive	365 (9.2)	3 (4.5)	362 (9.3)	
Klebsiella pneumoniae	254 (69.6)	2 (66.7)	252 (9.6)	0,999[b]
Escherichia coli	50 (13.7)	-	50 (13.8)	
Escherichia coli + *Klebsiella pneumoniae*	22 (5.9)	1 (33.3)	21 (5.8)	
Enterobacter cloacae	8 (2.2)	-	8 (2.2)	
Klebsiella oxytoca	5 (1.4)	-	5 (1.4)	
Enterobacter hormaechei	4 (1.1)	-	4 (1.1)	
Klebsiella aerogenes	4 (1.1)	-	4 (1.1)	
Klebsiella pneumoniae + *Proteus mirabilis*	3 (0.9)	-	3 (0.8)	
Enterobacter kobei	2 (0.5)	-	2 (0.6)	
Proteus mirabilis	2 (0.5)	-	2 (0.6)	
Serratia marcescens	2 (0.5)	-	2 (0.6)	
Aeromonas hydrophila	1 (0.3)	-	1 (0.3)	
Citrobacter koseri	1 (0.3)	-	1 (0.3)	
Raoultella ornithinolytica	1 (0.3)	-	1 (0.3)	
Enterobacter kobei + *Escherichia coli*	1 (0.3)	-	1 (0.3)	
Escherichia coli + *Klebsiella oxytica*	1 (0.3)	-	1 (0.3)	
Klebsiella aerogenes + *Klebsiella pneumoniae*	1 (0.3)	-	1 (0.3)	
Klebsiella pneumoniae + *Enterobacter cloacae*	1 (0.3)	-	1 (0.3)	
Klebsiella voricola + *Escherichia coli*	1 (0.3)	-	1 (0.3)	
Klebsiella pneumoniae + *Escherichia coli* + *Citrobacter freundii*	1 (0.3)	-	1 (0.3)	
CRE test results by PCR	All patients (*n* = 500,) *n*(%)[a]	Upon admission to the hospital (*n* = 64) *n*(%)[a]	During hospitalization (*n* = 436) *n*(%)[a]	p
Negative	419 (83.8)	56 (87.5)	363 (83.3)	0.254[b]
Positive	81 (16.2)	8 (12.5)	73 (16.7)	
bla_{KPC}	5 (6.1)	-	5 (1.1)	1.000[b]
bla_{NDM}	17 (21.0)	1 (1.6)	16 (3.7)	0.710[b]
bla_{OXA-48}	68 (84.0)	7 (10.9)	61 (14.0)	0.696[b]
bla_{VIM}/bla_{IMP}	2 (2.5)	-	2 (0.5)	1.000[b]
bla_{KPC} + bla_{OXA-48}	1	-	1	
bla_{NDM} + bla_{OXA-48}	5	-	5	
bla_{KPC} + bla_{OXA-48} + bla_{NDM}	3	-	3	

[a] Column percentage

[b] Fisher's Exact test Upon admission to the hospital and During hospitalization were compared)

were positive together, In five patients, the "bla_{NDM}" and "bla_{OXA-48}" genes were positive together. In three patients, the bla_{KPC}, bla_{NDM} and bla_{OXA-48} genes were positive together (Table 3).

There were 31 patients whose CHROMAgar and PCR test samples were taken simultaneously and had CRE growth. *Klebsiella pneumoniae* grew in 21 of these patients; bla_{KPC} was found in 19%; bla_{NDM} was found in 28.6%; bla_{OXA-48} gene was found to be positive in 66.7%. *Escherichia coli* grew in seven patients; bla_{KPC} was found in 14.3%; bla_{NDM} was found in 14.3%; bla_{OXA-48} was found in 71.4% and bla_{VIM}/bla_{IMP} gene was found to be positive in 14.3%. "*Klebsiella variicola* and *Escherichia coli*" grew together in one patient. The bla_{OXA-48} gene was positive in one patient with co-growth of "*Escherichia coli* and *Klebsiella pneumoniae*" and in one patient with growth of *Klebsiella oxytoca*. No statistically significant relationship was found between

Table 4 Analysis of CRE-produced enterobacteria and PCR results with simultaneously taken CHROMAgar test

Enterobacteriaceae **grown in CHROMAgar test**	**Carbapenemase resistance in rectal swabs by PCR method**						
	***bla*$_{KPC}$** ***n* = 4** (%)[a] (%)[b]	***bla*$_{NDM}$** **n = 8** (%)[a] (%)[b]	***bla*$_{OXA-48}$** ***n* = 21** (%)[a] (%)[b]	***bla*$_{VIM}$/*bla*$_{IMP}$** ***n* = 1** (%)[a] (%)[b]	***bla*$_{KPC}$** **+** ***bla*$_{OXA-48}$** ***n* = 1** (%)[a] (%)[b]	***bla*$_{NDM}$** **+** ***bla*$_{OXA-48}$** ***n* = 1** (%)[a] (%)[b]	***bla*$_{KPC}$** **+** ***bla*$_{OXA-48}$** **+** ***bla*$_{NDM}$** ***n* = 2** (%)[a] (%)[b]
Klebsiella pneumoniae (n = 21)	4 (19.0) (100.0)	6 (28.6) (75.0)	14 (66.7) (66.6)	-	*1 (4.8)* (100.0)	*2 (9.5)* (100.0)	*2 (9.5)* (100.0)
Escherichia coli (n = 7)	-	1 (14.3) (12.5)	5 (71.4) (23.8)	1 (14.3) (100.0)	-	-	-
Klebsiella voricola + Escherichia coli (n = 1)	-	1 (100.0) (12.5)	-	-	-	-	-
Escherichia coli + Klebsiella pneumoniae (n = 1)	-	-	1 (100.0) (4.7)		-	-	-
Klebsiella oxytoca (n = 1)			1 (100.0) (4.7)		-	-	-
P[c]	0.701	0.387	0.540	0.471	-	-	-

[a] Row percentage

[b] Colon percentage

[c] Fisher's exact test

Table 5 Status of infection focus development in patients with positive carbapenem-resistant Enterobacteriaceae colonization, as determined using CHROMAgar and PCR methods

	N	**Carbapenem-resistant infection at any focus**	
		Infection(+) ***n*(%)**	**Infection (-)** ***n*(%)[a]**
CRE positive patients by CHROMAgar	365	159 (43.6)	206 (56.4)
CRE positive patient by PCR method	81	16 (19.8)	65 (80.2)
CRE positive patient by PCR method + by CHROMAgar	31	16 (51.6)	15 (48.3)

CRE-producing bacteria and gene distribution ($p > 0.05$) (Table 4).

In the rectal swab samples of all 3953 patients, there were 365 CRE positive patients with CHROMAgar culture method, 81 with PCR and 31 with both methods. The total number of CRE positives was 384. The CRE positivity rate was found to be 9.7% in all these patients.

Table 5 presents the comparative results of patients with positive CRE colonization in CHROMAgar and PCR methods who developed infection in any focus. The data in that table reveal that the rate of developing infection during hospitalization was 19.8% in those with positive CRE detection only by PCR method and 43.6% in those with positive CRE detection only by CHROMAgar.

Discussion

Identifying and isolating CPE carriers among hospitalized patients is important to prevent the spread of serious infections, especially in hospital settings [15, 28]. Phenotypic tests show limitations such as low sensitivity and/or specificity and/or time consumption and/or difficulty in interpretation and cannot detect all carbapenemase types [19, 29]. Rapidly detecting CRE carriers is crucial because it has important clinical implications, such as implementation of infection control measures [23].

In our study, CRE screening was performed in rectal swabs of 3593 patients over a 2-year period. CRE colonization was detected in 9.7% patients with both methods. Rectal swab samples were sent mostly from intensive care units and CRE positivity was seen mostly in these groups. It was found to be statistically significant ($p < 0.05$). Many studies investigating the frequency of CRE colonization reported that it was most common in intensive care units, drew attention to risk factors, and strictly implemented infection control measures were mentioned [23]. Yamamoto et al. [24] reported 12.2% CRE carriage (in stool sample screening) in hospitalized patients. Zhao et al. [25] determined a stool carriage rate of 2.6% for CRE among hospitalized patients in China. Huang et al. [30] defined the CRE rate as 9.6% on average in their multicenter study. There are studies from Turkey reporting CRE colonization rates and high risk of spread [31–35]. Some of these studies; Zarakoğlu et al. [34] reported CRE colonization as 6.8% in patients in a university hospital, Perçin et al. [35] reported CRE carriage as 2.5% in rectal

swab screening in patients. These studies have shown that CRE colonization in rectal swabs may have different carriage rates in each region, center and even clinic. This difference may depend on the characteristics of the inpatients in the clinics, the infection control measures implemented and antibiotic use policies.

In our study, CRE carrier status was detected in 9.2% with CHROMAgar and in 16.2% with PCR screening test. Although the PCR test was studied in fewer patients than the CHROMAgar test number, the CRE colonization rate is higher. When the reason for this was examined, it was found that PCR screening test was only allowed to be used in special patients such as organ transplantation units in our hospital. This may be due to the higher number of hospital stays in the patient group in whom rectal CRE screening test with PCR was requested and the possibility of more predisposing factors such as antibiotic use and steroid use. In addition, in the screening test performed with the culture method, beta-lactamase producing microorganisms such as *Pseudomonas aeruginosa*, *Pseudomonas putida*, *Acinetobacter baumannii*, which grow outside the *Enterobacteriaceae* family, were detected but were not included in our records. The fact that gene positivity for these Gram negatives was detected in our PCR test may have increased the CRE positivity rate.

In our study, the most common microorganism showing CRE colonization detected in rectal swab samples was *Klebsiella pneumoniae* (69.6%), followed by *Escherichia coli* (13.7%), and *Enterobacter cloacae* (2.2%). Others were *Klebsiella oxytoca* (1.4%), *Enterobacter hormaechei*, *Klebsiella aerogenes*, *Enterobacter kobei* (1.1%), *Proteus mirabilis, Serratia marcescens, Aeromonas hydrophila*, *Citrobacter koseri, Raoultella ornithinolytica* (0.5%). *Enterobacteriaceae* species with high CRE frequency were found to be *K. pneumoniae* and *E. coli*, similar to many studies conducted in Türkiye and abroad [2, 35–38].

In our study, carbapenemase gene screening by PCR was performed on 500 patients. CPE gene positivity was found in 81 of these patients (16.2%). The "$bla_{\text{OXA-48}}$" gene was the most prevalent, with a rate of 84%, followed by "bla_{NDM}" (21.0%), "bla_{KPC}" (6.1%) and "$bla_{\text{VIM}/\text{IMP}}$" (2.5%) genes. Although the most common carbapenemase in our country is OXA-48, different carbapenemases have also been reported [36–38]. In this study, similar to the studies conducted in our country, the most common gene was "$bla_{\text{OXA-48}}$". In addition, "bla_{KPC}" and "$bla_{\text{OXA-48}}$" genes were positive in four patients, and "bla_{NDM}" and "$bla_{\text{OXA-48}}$" genes were positive together. There are studies in our country showing that these two genes were detected in a single strain, and similar reports abroad [39–41].

When the genes detected in PCR screening with Enterobacteriaceae growing with CHROMAgar in the same patient were examined; The frequency of all genes; bla_{KPC} was seen only in *K. pneumoniae* with 100%. $bla_{\text{OXA-48}}$; 66.6%, 23.8%, bla_{NDM}; 75%, 12.5% were seen in *K. pneumoniae and E. coli*, respectively. When the distribution of genes among bacteria was evaluated statistically, it was not found to be significant ($p > 0.05$). The rates detected in our study were similar to the studies conducted in our country and the $bla_{\text{OXA-48}}$ gene was most frequently identified in *K. pneumoniae* [22, 42].

Interestingly, in our study, we had three patients in whom bla_{KPC}, bla_{NDM} and $bla_{\text{OXA-48}}$ genes, which have not been reported before in İstanbul/Turkiye, were detected in a single strain (two *K. pneumoniae*, the other one was not identified as a microorganism). One of these patients was a general intensive care patient and the other was a hepatobiliary surgery clinic patient due to a cancer diagnosis. There was no growth in the CHROMAgar culture of one patient but these three genes were detected by PCR and he was a general intensive care patient. Carbapenem-resistant *K. pneumoniae* was grown in the blood culture during the hospital stay. The presence of these 3 genes in a single strain may cause increased concerns. Because the co-production of carbapenemases from different classes is clinically and epidemiologically important and can cause multidrug resistance in infections and infections are associated with high mortality rates [42, 43]. There are several studies reporting the co-occurrence of these three genes in our country and abroad [44–46]. In addition, there has been a very intense migration to Istanbul since 2011 due to war and socioeconomic reasons. Therefore, our hospital is a large hospital that accepts foreign patients and undertakes their diagnosis and treatment. It is also possible that combinations of carbapenemase-producing genes may emerge through patients coming from different geographical regions.

Given that CRE colonization is an independent risk factor for CRE infection, asymptomatic CRE carriers are generally at risk for subsequent CRE infections [21]. Another aim of this study was to investigate whether meropenem-resistant infection developed among CRE carrier patients in our hospital from the day of hospitalization to the day of discharge and A meropenem-resistant infection focus was detected in 39.2% of CRE colonization-positive patients. n a study from Jordan, the incidence of CRE infections was 41.3% [47], in a systematic review analyzing CRE colonized patients, the cumulative rate of subsequent infections was 16.5% [48] and another study showed that 36.09% of CRE colonizations progressed to infection [49]. In our study, although it is difficult to say that the meropenem-resistant CRE

infection developed in the patients directly developed from their own colonized bacteria, the rate found is similar to other studies. When we compared the rates of meropenem-resistant infection developing in CRE positive patients with PCR and CHROMAgar methods, they were 19.8% and 43.6%, respectively. The rate of developing carbapenem-resistant infection was found to be lower in patients diagnosed with PCR method. This result may have been obtained due to reasons such as rapid diagnosis with the PCR method, ensuring patient isolation, and stricter application of infection control measures by patient care personnel. In a study where cost-effectiveness studies were conducted, it was reported that early diagnosis with PCR was more effective [50]. It was thought that the most appropriate models for CRE screening tests should be performed in our center in order to identify patients at high risk of infection among CPE carriers and to facilitate the establishment of evidence-based management guidelines.

As a result, the frequency of CRE rectal colonization in our hospital is 9.7%. It was found at a high rate in intensive care units and was evaluated as statistically significant ($p = 0.05$). This rate is higher than many centers. The most common isolated microorganisms from the CRE were *K. pneumoniae* and the second most common was *E. coli*. Among the resistance genes, the $bla_{\text{OXA-48}}$ gene, which is the most frequently identified gene in Turkiye, is also the most frequently seen gene in our center, and the second most frequently seen gene was $bla_{\text{KPC.}}$ Unlike many centers, multiple genes were detected in a single bacterium in our hospital. Most strikingly, CRE colonizations containing the $bla_{\text{KPC.}} + bla_{\text{OXA-48}} + bla_{\text{NDM}}$ genes in a single strain were detected in three separate patients. In our hospital, infection control measures need to be reviewed urgently or it is recommended to monitor how much attention is paid to infection control measures by the staff. During hospitalization, the development of carbapenem-resistant infections at various focuses was higher in patients diagnosed with CRE colonization with CHROMAgar than in patients diagnosed with PCR.

Limitations

The study has all the limitations of a retrospective single-center study. CRE screening test with PCR is not a routine practice for all hospitalized patients in our hospital. It is routinely applied in the liver and biliary tract diseases unit and transplantation unit, Therefore, our results could only be compared with 500 patients for whom PCR was requested. CRE colonization test was not requested for all patients at the time of admission to the hospital. Patients in whom CRE colonization was not detected were not followed up regularly. For this reason, it could not be determined on which day of hospitalization the CRE positivity was positive. Rectal screening test was not performed on some CRE colonization-positive patients at the time of admission to the hospital. Therefore, it could not be distinguished whether the patients with CRE carrier status were actually positive at the time of admission to the hospital or became positive after hospitalization. Since it was a retrospective study, predisposing effects could not be learned because the patients' past disease history, antibiotic use during hospitalization, and other reasons could not be taken into account. Since the past history of the outpatients was unknown, it could not be defined whether it was community-acquired or not. In addition, a comparison using molecular methods could not be made to determine whether the Enterobacteriaceae produced in the carbapenem-resistant infection foci that developed in the patients were the same as the Enterobacteriaceae species detected in their CRE colonization.

Conclusions

Although CRE colonization was found to be higher in the patient group where the PCR method was applied, the low development of carbapenem-resistant infection foci shows the importance of early diagnosis and isolation. In addition, the co-existence of 3 CRE genes ($bla_{\text{KPC}} + bla_{\text{NDM}} + bla_{\text{OXA-48}}$) in 3 patients two of whom was identified as *K. pneumoniae*) may cause multidrug-resistant outbreaks in clinics, considering that resistance genes can be transferred to other Gram-negative bacilli, including *Enterobacteriaceae*. Our study has shown that cost-effectiveness analyses should be performed on rectal swab samples and algorithms for effective and rapid diagnostic methods should be developed.

Abbreviations

CPE	Carbapenemase-Producing *Enterobacteriaceae*
CRE	Carbapenem-Resistant *Enterobacteriaceae*
MALDI-TOF MS	Matrix-Assisted Laser Desorption Ionization Time-of-Flight Mass Spectrometry
K. pneumoniae	*Klebsiella pneumoniae*
E. coli	*Eschericha coli*
EUCAST	The European Committee on Antimicrobial Susceptibility Testing
CDC	Centers for Disease Control and Prevention
PCR	Polymerase chain reaction

Authors' contributions

K.Ş. designed the study, supervised the data collection process, and cleaned the data. B.Ö. helped with the data cleaning and analysis and prepared the final manuscript. K.Ş., B.Ö. have read and agreed to the published version of the manuscript.

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Data availability
The datasets generated and/or analyzed during the current study are not publicly available due to original data rights belonging to the main study but are available from the corresponding author on reasonable request pending approval from the main study principal investigator.

Declarations

This study was conducted in accordance with the principles of the Declaration of Helsinki.Ethics approval and consent to participate
This study was approved by the Çam and Sakura City Hospital Ethics Committee (Approval Number: [26/06/2024 No. KAEK/12.06.2024.36.]. The need for informed consent was waived by the Çam and Sakura City Hospital Ethics Committee, as the study was designed as a retrospective analysis and involved no direct contact with patients.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

Author details
[1]University of Health Science, Başakşehir Çam and Sakura City Hospital, Medical Microbiyology, Başakşehir, Istanbul 34480, Turkey.

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References

1. Janda JM, Abbott SL. The changing face of the family *Enterobacteriaceae* (order: "*Enterobacterales*"): new members, taxonomic issues, geographic expansion, and new diseases and disease syndromes. Clin Microbiol Rev. 2021;34:10.1128/cmr.00174-20. https://doi.org/10.1128/cmr.00174-20.
2. Tilahun M, Kassa Y, Gedefie A, Ashagire M. Emerging carbapenem-resistant Enterobacteriaceae infection, its epidemiology and novel treatment options: a review. Infect Drug Resist. 2021;14:4363–74. https://doi.org/10.2147/IDR.S337611. PMID: 34707380; PMCID: PMC8544126.
3. Nordmann P, Dortet L, Poirel L. Carbapenem resistance in Enterobacteriaceae: here is the storm! Trends Mol Med. 2012;18:263–72. https://doi.org/10.1016/j.molmed.2012.03.003. PMID: 22480775.
4. Gijón D, Curiao T, Baquero F, Coque TM, Cantón R. Fecal carriage of carbapenemase-producing Enterobacteriaceae: a hidden reservoir in hospitalized and nonhospitalized patients. J Clin Microbiol. 2012;50(5):1558–63. https://doi.org/10.1128/JCM.00020-12. Epub 2012 Mar 7. PMID: 22403422; PMCID: PMC3347124.
5. Munoz-Price LS, Poirel L, Bonomo RA, et al. Clinical epidemiology of the global expansion of Klebsiella pneumoniae carbapenemases. Lancet Infect Dis. 2013;13:785–96. https://doi.org/10.1016/S1473-3099(13)70190-7. PMID: 23969216; PMCID: PMC4673667.
6. Epson EE, Pisney LM, Wendt JM, et al. Carbapenem-resistant Klebsiella pneumoniae producing New Delhi metallo-β-lactamase at an acute care hospital, Colorado, 2012. Infect Control Hosp Epidemiol. 2014;35:390–7. https://doi.org/10.1086/675607. PMID: 24602944.
7. Epstein L, Hunter JC, Arwady MA, et al. New Delhi metallo-β-lactamase-producing carbapenem-resistant Escherichia coli associated with exposure to duodenoscopes. JAMA. 2014;312:1447–55. https://doi.org/10.1001/jama.2014.12720.
8. Foschi C, Gaibani P, Lombardo D, Re MC, Ambretti S. Rectal screening for carbapenemase-producing Enterobacteriaceae: a proposed workflow. J Glob Antimicrob Resist. 2020;21:86–90. https://doi.org/10.1016/j.jgar.2019.10.012. Epub 2019 Oct 19 PMID: 31639545.
9. Tzouvelekis LS, Markogiannakis A, Psichogiou M, Tassios PT, Daikos GL. Carbapenemases in Klebsiella pneumoniae and other Enterobacteriaceae: an evolving crisis of global dimensions. Clin Microbiol Rev. 2012;25(4):682–707. https://doi.org/10.1128/CMR.05035-11. PMID: 23034326; PMCID: PMC3485753.
10. Daikos GL, Tsaousi S, Tzouvelekis LS, et al. Carbapenemase-producing Klebsiella pneumoniae bloodstream infections: lowering mortality by antibiotic combination schemes and the role of carbapenems. Antimicrob Agents Chemother. 2014;58(4):2322–8. https://doi.org/10.1128/AAC.02166-13. Epub 2014 Feb 10. PMID: 24514083; PMCID: PMC4023796.
11. Monaco M, Giani T, Raffone M. et al. Colistin resistance superimposed to endemic carbapenem-resistant Klebsiella pneumoniae: a rapidly evolving problem in Italy, November 2013 to April 2014. Euro Surveill. 2014;19(42). https://doi.org/10.2807/1560-7917.es2014.19.42.20939. PMID: 25358041.
12. Akova M, Daikos GL, Tzouvelekis L, Carmeli Y. Interventional strategies and current clinical experience with carbapenemase-producing Gram-negative bacteria. Clin Microbiol Infect. 2012;18(5):439–48. https://doi.org/10.1111/j.1469-0691.2012.03823.x. PMID: 22507111.
13. De Rosa FG, Corcione S, Cavallo R, Di Perri G, Bassetti M. Critical issues for Klebsiella pneumoniae KPC-carbapenemase producing K. pneumoniae infections: a critical agenda. Future Microbiol. 2015;10(2):283–94. https://doi.org/10.2217/fmb.14.121. PMID: 25689539.
14. Grundmann H, Livermore DM, Giske CG, et al. Carbapenem-non-susceptible Enterobacteriaceae in Europe: conclusions from a meeting of national experts. Euro Surveill. 2010;15(46). https://doi.org/10.2807/ese.15.46.19711. PMID: 21144429.
15. Lledo W, Hernandez M, Lopez E, et al. Guidance for control of infections with carbapenem-resistant or carbapenemase-producing Enterobacteriaceae in acute care facilities. MMWR Morb Mortal Wkly Rep. 2009;58(10):256. https://www.cabidigitallibrary.org/doi/full/10.5555/20093106836.
16. Landman D, Salvani J, Bratu S, Quale J. Evaluation of techniques for detection of carbapenem-resistant Klebsiella pneumoniae in stool surveillance cultures. J Clin Microbiol. 2005;43(11):5639–41. https://doi.org/10.1128/JCM.43.11.5639-5641.2005. PMID: 16272497; PMCID: PMC1287836.
17. Cohen Stuart J, Leverstein-Van Hall MA. Guideline for phenotypic screening and confirmation of carbapenemases in Enterobacteriaceae. Int J Antimicrob Agents. 2010;36(3):205–10. https://doi.org/10.1016/j.ijantimicag.2010.05.014. Epub 2010 Jul 3. PMID: 20598859.
18. Panagea T, Galani I, Souli M, Adamou P, Antoniadou A, Giamarellou H. Evaluation of CHROMagar™ KPC for the detection of carbapenemase-producing Enterobacteriaceae in rectal surveillance cultures. Int J Antimicrob Agents. 2011;37(2):124–8. https://doi.org/10.1016/j.ijantimicag.2010.10.010. Epub 2010 Dec 21 PMID: 21177079.
19. Saliba R, Aho-Glélé LS, Karam-Sarkis D, Zahar JR. Evaluation of polymerase chain reaction assays for direct screening of carbapenemase-producing Enterobacteriaceae from rectal swabs: a diagnostic meta-analysis. J Hosp Infect. 2020;104(3):381–9. https://doi.org/10.1016/j.jhin.2019.11.017. Epub 2019 Nov 29 PMID: 31790743.
20. Viale P, Tumietto F, Giannella M, et al. Impact of a hospital-wide multifaceted programme for reducing carbapenem-resistant Enterobacteriaceae infections in a large teaching hospital in northern Italy. Clin Microbiol Infect. 2015;21(3):242–7. https://doi.org/10.1016/j.cmi.2014.10.020. Epub 2014 Nov 11 PMID: 25658534.
21. Solgi H, Badmasti F, Aminzadeh Z, et al. Molecular characterization of intestinal carriage of carbapenem-resistant Enterobacteriaceae among inpatients at two Iranian university hospitals: first report of co-production of bla (NDM-7) and bla (OXA-48). Eur J Clin Microbiol Infect Dis. 2017;36(11):2127–35. https://doi.org/10.1007/s10096-017-3035-3. Epub 2017 Jun 21 PMID: 28639165.
22. Liu J, Zhang H, Feng D et al. Development of a risk prediction model of subsequent bloodstream infection after carbapenem-resistant Enterobacteriaceae isolated from perianal swabs in hematological patients. Infect Drug Resist. 2023:1297–312. https://doi.org/10.2147/IDR.S400939. PMID: 36910516; PMCID: PMC9999719.
23. Al Fadhli AH, Jamal WY, Rotimi VO. Prevalence of carbapenem-resistant Enterobacteriaceae and emergence of high rectal colonization rates of blaOXA-181-positive isolates in patients admitted to two major hospital intensive care units in Kuwait. PLoS ONE. 2020;15(11):e0241971. https://doi.org/10.1371/journal.pone.0241971.
24. Yamamoto N, Asada R, Kawahara R, et al. Prevalence of, and risk factors for, carriage of carbapenem-resistant Enterobacteriaceae among hospitalized patients in Japan. J Hosp Infect. 2017;97(3):212–7. https://doi.org/10.1016/j.jhin.2017.07.015. Epub 2017 Jul 20 PMID: 28736270.
25. Zhao ZC, Xu XH, Liu MB, Wu J, Lin J, Li B. Fecal carriage of carbapenem-resistant Enterobacteriaceae in a Chinese university hospital. Am J Infect

Control. 2014;42(5):e61–4. https://doi.org/10.1016/j.ajic.2014.01.024. PMID: 24773806.
26. Righi E, et al. ESCMID/EUCIC clinical practice guidelines on perioperative antibiotic prophylaxis in patients colonized by multidrug-resistant Gram-negative bacteria before surgery. Clin Microbiol Infect. 2023;29(4):463–79.
27. https://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Breakpoint_tables/v_15.0_Breakpoint_Tables.pdf
28. Ben-David D, Maor Y, Keller N, et al. Potential role of active surveillance in the control of a hospital-wide outbreak of carbapenem-resistant Klebsiella pneumoniae infection. Infect Control Hosp Epidemiol. 2010;31(6):620–6. https://doi.org/10.1086/652528. PMID: 20370465.
29. Dortet L, Bréchard L, Cuzon G, Poirel L, Nordmann P. Strategy for rapid detection of carbapenemase-producing Enterobacteriaceae. Antimicrob Agents Chemother. 2014;58(4):2441–5. https://doi.org/10.1128/AAC.01239-13. Epub 2014 Jan 27. PMID: 24468779; PMCID: PMC4023735.
30. Huang TD, Bogaerts P, Ghilani E, et al. Multicentre evaluation of the Check-Direct CPE® assay for direct screening of carbapenemase-producing Enterobacteriaceae from rectal swabs. J Antimicrob Chemother. 2015;70(6):1669–73. https://doi.org/10.1093/jac/dkv009. Epub 2015 Jan 29 PMID: 25637518.
31. Nordmann P, Naas T, Poirel L. Global spread of carbapenemase-producing Enterobacteriaceae. Emerg Infect Dis. 2011;17(10):1791–8. https://doi.org/10.3201/eid1710.110655. PMID: 22000347; PMCID: PMC3310682.
32. Yalcinkaya A, Öztaş YE, Sabuncuoğlu S. Sterols in inflammatory diseases: implications and clinical utility. In: Lizard G, editor. Implication of oxysterols and phytosterols in aging and human diseases. Cham: Springer International Publishing; 2024. p. 261–75. https://doi.org/10.1007/978-3-031-43883-7_13. PMID: 38036884.
33. Fauconnier C, Dodemont M, Depouhon A, Anantharajah A, Verroken A, Rodriguez-Villalobos H. Lateral flow immunochromatographic assay for rapid screening of faecal carriage of carbapenemase-producing Enterobacteriaceae. J Antimicrob Chemother. 2019;74(2):357–9. https://doi.org/10.1093/jac/dky429. PMID:30376099.az.
34. Zarakolu P, Eser OK, Aladag E, Al-Zahrani IA, Day KM, Atmaca O, Boral B, Cakir B, Perry JD, Akova M. Epidemiology of carbapenem-resistant Klebsiella pneumoniae colonization: a surveillance study at a Turkish university hospital from 2009 to 2013. Diagn Microbiol Infect Dis. 2016;85(4):466–70. https://doi.org/10.1016/j.diagmicrobio.2016.05.012. Epub 2016 May 21 PMID: 27306118.
35. Perçin D, Colakoğlu S, Durmaz S, Ekincioğlu P. Comparison of ertapenem-EMB Agar with traditional methods for screening carbapenem-resistant Klebsiella pneumoniae from rectal swabs. Mikrobiyoloji Bulteni. 2012;46(4):546–52. PMID: 23188568.
36. Alizadeh N, Ahangarzadeh-Rezaee M, et al. Evaluation of resistance mechanisms in carbapenem-resistant Enterobacteriaceae. Infect Drug Resist. 2020;13:1377–85. https://doi.org/10.2147/IDR.S244357. PMID: 32494169; PMCID: PMC7229782.
37. Bush K. Alarming β-lactamase-mediated resistance in multidrug-resistant Enterobacteriaceae. Curr Opin Microbiol. 2010;13(5):558–64. https://doi.org/10.1016/j.mib.2010.09.006. Epub 2010 Oct 1 PMID: 20920882.
38. Gülmez D, Woodford N, Palepou M-FI, et al. Carbapenem-resistant Escherichia coli and Klebsiella pneumoniae isolates from Turkey with OXA-48-like carbapenemases and outer membrane protein loss. Int J Antimicrob Agents. 2008;31(6):523–6. https://doi.org/10.1016/j.ijantimicag.2008.01.017. Epub 2008 Mar 12 PMID: 18339523.
39. Raheel A, Azab H, Hessam W, Abbadi S, Ezzat A. Detection of carbapenemase enzymes and genes among carbapenem-resistant Enterobacteriaceae isolates in Suez Canal University Hospitals in Ismailia, Egypt. Microbes Infect Dis. 2020;1(1):24–33. https://doi.org/10.21608/mid.2020.25702.1007.
40. Okalin ŞŞ, Sarı- Kaygısız AN, Ergon MC, Öktem İMA. Investigation of carbapenemase genes in carbapenem resistant Enterobacterales isolates: first kpc report from Dokuz Eylul University Hospital. Turk Mikrobiyol Cemiy Derg. 2021;51(4):375–81. https://doi.org/10.5222/TMCD.2021.94899. https://tmc.dergisi.org/pdf/pdf_TMC_747.pdf.
41. Otlu B, Yakupoğulları Y, Gürsoy NC, Duman Y, Bayındır Y, Tekerekoğlu MY, Ersoy M. CoProduction of OXA-48 and NDM-1 carbapenemases in Providencia rettgeri: the first report. Mikrobiyol Bul. 2018;52(3):300–7. https://www.mikrobiyolbul.org/managete/fu_folder/2018-03/2018-52-3-300-307.pdf.
42. Cantón R, Akóva M, Carmeli Y, et al. Rapid evolution and spread of carbapenemases among Enterobacteriaceae in Europe. Clin Microbiol Infect. 2012;18(5):413–31. https://doi.org/10.1111/j.1469-0691.2012.03821.x. PMID: 22507109.
43. Gülay Z. Molecular epidemiology of carbapenemases in Enterobacteriaceae. ANKEM Derg. 2014;28(Ek 2):73–76. https://ankemdernegi.org.tr/ANKEMJOURNALPDF/ANKEM_28_Ek2_73_76.pdf.
44. Ishahrani AM, Ibrahim ME, Aldossary AK, Alghamdi MA, Ahmed OB, Bin Abdulhak AA. Molecular epidemiology of carbapenem-resistant *K pneumoniae* clinical isolates from the adult patients with comorbidities in a tertiary hospital, *Southern Saudi Arabia*. Antibiotics (Basel). 2022;11(12):1697. https://doi.org/10.3390/antibiotics11121697. PMID: 36551354; PMCID: PMC9774885.
45. Xie L, Dou Y, Zhou K, Chen Y, Han L, Guo X, Sun J. Coexistence of blaOXA−48 and truncated blaNDM−1 on different plasmids in a Klebsiella pneumoniae isolate in China. Front Microbiol. 2017;8:133. https://doi.org/10.3389/fmicb.2017.00133. PMID: 28210248; PMCID: PMC5288367.
46. Alkan B, Bayraktar Ö, Özcan N, Gül K, Akpolat N. Dissemination of blaOXA-48 like, blaNDM, blaKPC, blaIMP-1, blaVIM genes among carbapenem-resistant Escherichia coli and Klebsiella pneumoniae strains in Southeastern Turkey: first report of Klebsiella pneumoniae co-producing blaOXA-48-like, blaVIM and blaIMP-1 genes. Rev Med Microbiol. 2021;32(4):205–10. https://doi.org/10.1097/MRM.0000000000000287.
47. Hammour KA, Abu-Farha R, Itani R, et al. The prevalence of carbapenem resistance gram negative pathogens in a Tertiary Teaching Hospital in Jordan. BMC Infect Dis. 2023;23:634. https://doi.org/10.1186/s12879-023-08610-4.
48. Tischendorf J, de Avila RA, Safdar N. Risk of infection following colonization with carbapenem-resistant Enterobactericeae: a systematic review. Am J Infect Control. 2016;44(5):539–43.
49. Gao Y, Chen M, Cai M, et al. An analysis of risk factors for carbapenem-resistant Enterobacteriaceae infection. J Glob Antimicrob Resist. 2022;30:191–8. https://doi.org/10.1016/j.jgar.2022.04.005. PMID: 35429666.
50. Moloney E, Lee KW, Craig D, Allen AJ, Graziadio S, Power M, Steeds CA. PCR-based diagnostic testing strategy to identify carbapenemase-producing Enterobacteriaceae carriers upon admission to UK hospitals: early economic modelling to assess costs and consequences. Diagn Progn Res. 2019;18(3):8. https://doi.org/10.1186/s41512-019-0053-x. PMID: 31093578; PMCID: PMC6471810.

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