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Original Article

Enterobacterial infection in Saudi Arabia: First record of *Klebsiella pneumoniae* with triple carbapenemase genes resistance

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Abstract

Introduction: Carbapenemase producing Enterobacteriaceae are emerging as important pathogens worldwide with serious effects on patients' outcome. The study aimed to investigate the emergence of carbapenemases associated with enterobacterial infection in Western region of Saudi Arabia.

Methodology: Clinical isolates from suspected patients with enterobacterial infection were investigated over a one-year period from four tertiary care hospitals of Makkah, Saudi Arabia. All isolates were identified using Vitek-2 system and then screened for potential carbapenemase production using disk diffusion test. Suspected isolates with reduced susceptibility to carbapenems were further investigated for bla_{NDM-1}, bla_{KPC} and bla_{OXA-48} resistant genes.

Results: Out of 120 confirmed Enterobacteriaceae isolates, Klebsiella pneumoniae and Escherichia coli comprised the largest proportion (35% and 34.2%, respectively) of encountered infections. Twenty-six (21.7%) isolates showed resistance to carbapenems, the majority of which (21/26) were K. pneumoniae. Remarkably, 17 isolates carried triple resistant genes KPC/NDM-1/OXA-48 while the other 4 carried double resistant genes (KPC/OXA-48) or (NDM-1/OXA-48). The current study revealed that the mentioned triple resistance genes have the higher incidence with significant association risk among males (COR 4.5; CI: 1.9-17.3; P = 0.018), non-Saudi nationalities (COR 4.9; CI: 1.5-19.3; P = 0.003), ICU-obtained specimens (COR 3.6; CI: 1.5-8.4; P = 0.002) and blood specimens (COR 2.8; CI: 1.1-6.9; P = 0.02).

Conclusion: Multidrug-resistant Enterobacteriaceae isolates in particular K. pneumoniae co-harboring KPC, NDM-1 and OXA-48 genes are emerging in Western region, Saudi Arabia. This is the first record of triple carbapenemase genes co-producing K. pneumoniae associated with enterobacterial infection.

Key words: Enterobacterial infection; *K. pneumoniae*; carbapenemase genes; KPC; NDM-1; OXA-48.

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Introduction

There has been emergence of isolates producing carbapenemases that efficiently hydrolyze carbapenems as well as most β-lactam drugs. The most common carbapenemases reported worldwide among *Enterobacteriaceae* are the Ambler class A *Klebsiella pneumoniae* carbapenemase (KPC), class B metallo-β-lactamases (VIM, IMP, NDM), and class D (OXA-48) types. These are most commonly found in *K. pneumoniae* isolates that are frequently associated with nosocomial infections and outbreaks [1].

The prevalence of carbapenemases-producing *Enterobacteriaceae* (KPC, NDM, OXA-48) varies significantly among different countries. The KPC has been reported worldwide but more predominantly found in the United States, Greece, South America, China and Taiwan [2]. Similarly, most of the NDM cases indicated a link with the Indian subcontinent [3,4] or Balkan countries [5,6] and from Middle East region [7-11]. Whereas, the OXA-48 producing *K. pneumoniae* is endemic in Turkey, Morocco, Libya, Egypt, Tunisia [12] and has also been documented in Mediterranean region [13], Middle East [14-16] and European countries [17,18]. All three types of carbapenemases have been frequently described from India, Spain, France, Italy and United Kingdom [19]. In Arabian Peninsula, recent studies have shown the predominance of OXA-48 and NDM type carbapenemases [11,20] where both OXA-48 and NDM producing *K. pneumoniae* were found in Oman [7,15], Kuwait [8], Qatar [11] and Saudi Arabia [10,16]. Instead, the region used to be free from KPC-producing *Enterobacteriaceae* where no previous studies claimed their isolation. However, recently two *K. pneumoniae*

carrying bla_{KPC-2} were isolated from 2 local patients in United Arab Emirates (UAE) and denoted the emergence of KPC-producing strains for the first time in Arabian Peninsula [21].

Multi-drug resistant *K. pneumoniae* isolates usually carry a single carbapenemase (e.g., KPC, NDM, OXA-48-like), however, unique and rare *K. pneumoniae* isolates co-producing two classes of carbapenemases have also been reported from Singapore [22] and countries of Europe [23,24] and Middle East [15,25]. In Saudi Arabia, the emergence of NDM-1 or OXA-48 has also been reported in *K. pneumoniae* isolates, however, none of these isolates either harbored KPC gene or combination of NDM-1 and OXA-48 [16].

Considering the extensive links of Makkah to the rest of the world as a target for millions of people from all over the world, the aim of the current study was to assess the resistance profile among *Enterobacteriaceae* isolates and to investigate the emergence of KPC, NDM-1 and OXA-48 resistance genes in Makkah (Western region), Saudi Arabia.

Methodology

Study Design

In a cross-sectional study, different clinical specimens were collected from suspected patients admitted to different wards of the four main tertiary care hospitals of Makkah over a period of 1 year (January 2017 to December 2017). Inclusion criteria included any suspected patient with clinical symptoms related to respiratory tract, urinary tract, gastrointestinal tract, and wound infections. Specimens from patients on antibiotic therapy were excluded from the study. A total of 864 specimens were investigated for the causative agents belonging to *Enterobacteriaceae*. Recovered isolates were identified using Vitek 2 compact system (BioMerieux, USA) and then screened for reduced susceptibility to carbapenems and further investigated for the presence of carbapenemases genes. In addition to type of collected specimens and admission wards, demographic information of corresponding patients including age, gender and nationality were obtained using a predesigned patient data sheet.

The medical ethics committee of Umm Al Qura University has approved the current study in accordance with the declaration of Helsinki (AMSEC 7/10-12-2016). A written consent was collected from all participating patients acknowledging the inclusion of their data anonymously in the study.

Screening for reduced susceptibility to carbapenems

All recovered *Enterobacteriaceae* isolates were first screened for reduced susceptibility to carbapenems by disk diffusion method using ceftazidime (30µg), cefotaxime (30µg), cefpodoxime (10µg), imipenem (10µg) and meropenem (10µg) following guidelines of Clinical and Laboratory Standards Institute (CLSI) [26]. In addition, minimum inhibitory concentration (MIC) of isolates which showed reduced susceptibility or resistance to carbapenems in screening test were determined for ceftazidime, cefotaxime, ceftriaxone, cefepime, imipenem, meropenem, amikacin, gentamicin and colistin using Gram-negative antibiotic susceptibility panel of Vitek-2 system.

Detection of carbapenemases genes

All suspected isolates of carbapenem-resistant *Enterobacteriaceae* (CRE) were further tested for the production of carbapenemase genes by standard PCR method. Chromosomal DNA and/or bacterial plasmid DNA was extracted from selected bacterial strains using DNeasy Blood and Tissue kit (Qiagen, Dammam, Saudi Arabia) and/or PureYield Plasmid Miniprep System (Promega Corporaon, Madison, WI, USA), respectively according to manufacturer's instructions. The bla_{NDM-1} gene was detected by PCR amplification of a 758 bp-specific product using forward primer (5'-GGGCCGTATGAGTGA-3') and reverse primer (5'-GAAGCTGAGCACCGCATTAG-3') as described earlier by Sidjabat *et al*. [27]. The detection of bla_{KPC} gene was carried out using primer set KPC-1 (5'-ATGTCACTGTATCGCCGTC-3') and KPC-2 (5'-AATCCCTCGAGCGCGAGT-3') for generation of a 862 bp-specific product as previously described by Mulvey *et al.* [28], while primer set OXA-48A (5'-TTGGTGGCATCGATTATCGG-3') and OXA-48B (5'-GAGCACTTCTTTTGTGATGGC-3') was used for detection of bla_{OXA-48} gene by amplifying a 743 bp-specific product after Poirel *et al*. [29].

Statistical analysis

Statistical analysis of the results was performed using SPSS version 16 (SPSS, Chicago, IL, USA). The frequencies of CRE isolates and associated resistance genes were assessed based on gender, nationalities, type and source of specimens using cross-tabulation followed by Chi square (χ^2) test or Fischer's exact test. A crude odd ratio (COR) with 95% confidence interval (CI) was calculated for frequency analysis as appropriate for the assessment of possible association risk ratio between resistance genes and different factors including patient's nationality, gender, type and source

of infection. The tests were two tailed and P value < 0.05 was considered significant.

Results

Descriptive analysis of study population

A total of 120 cases out of 864 suspected patients were confirmed with *Enterobacteriaceae* infection and served as the study population. Descriptive analysis of study population is presented in Table 1 showing the frequency of investigated population in relation to gender and nationality of patients as well as type and source of collected specimens. The results revealed that the majority of *Enterobacteriaceae*-infected study population was of Saudi nationality (65.8%). In term of source and type of specimens, the majority of specimens were encountered from medical ward (46.7%) and urine specimens (44.2%), respectively.

Identification and resistance profile of Enterobacteriaceae isolates

Klebsiella pneumoniae and *Escherichia coli* together comprised 69.2 % of total 120 *Enterobacteriaceae* isolates. The remaining 30.8% included, *Serratia* species (5.8%), *K. ornithinolytica* and *Proteus mirabilis* (5%, each), *K. oxytoca* and *Enterobacter cloacae* (4.2%, each), *Citrobacter* species (3.3%), *Salmonella* species (2.5%) and *E.aerogenes* (0.8%). Twenty-six isolates were resistant to the tested cephalosporins (ceftazidime, cefotaxime, ceftriaxone, cefepime) in which the MICs of the most mentioned cephalosporins and tested carbapenems were between > 32-64 µg/mL and > 8-16 µg/mL, respectively. In addition, a high degree of resistance to gentamicin (> 8-16µg/mL) (65.3%) and amikacin (> 64µg/mL) (42.3%) were also recorded. No isolates were resistant to colistin except three strains of *P. mirabilis,* which are intrinsically resistant. The antibiotic susceptibility profile of CRE isolates in relation to organism, wards, specimens, nationality and associated carbapenemase genes (Table 1-S) and MICs of these isolates (Table 2-S) was determined. Antibiotic susceptibility profiles of all recovered enterobacterial isolates were also obtained (Table 3-S).

Frequency and distribution of carbapenem resistant Enterobacteriaceae (CRE)

Twenty-six out of 120 *Enterobacteriaceae* isolates were identified as carbapenem resistant. High significant difference (P < 0.001) in the frequency of carbapenem resistance was recorded among different *Enterobacteriaceae* species. Therefore, *K. pneumonia* was the most predominant strain among the others (80.8%) and that was followed by *E. coli* (7.7%), *E. cloacae* (7.7%) and *P. mirabilis* (3.8%) as the least strains (Table 2).

The distribution of CRE according to the nationality, gender, source and types of specimens showed that *K. pneumoniae* was slightly higher in non-Saudis (42.3%) compared to Saudis (38.5%). Other CRE such as *E. coli, E. cloacae* and *P. mirabilis* were

Table 1. Descriptive analysis of study population and specimens.

Population (n = 120)	**Age (years) (Min-Max) (Mean ± SD)**	**Nationality/Gender No (%)**			
		Saudi		**Non-Saudi**	
	(0.66 – 91.0) (24.6 ± 49.2)	79 (65.8)		41 (34.2)	
		Male	**Female**	**Male**	**Female**
		39 (49.4)	40 (50.6)	19 (46.3)	22 (53.7)
Specimens (n = 120)	**Source of specimens No (%)**	Intensive care unit		24 (20)	
		Medical ward		56 (46.7)	
		Surgical ward		23 (19.2)	
		Pediatric ward		12 (10.0)	
		Antenatal ward		1 (0.8)	
		Obstetrics and Gynecology		3 (2.5)	
		Outpatient		1 (0.8)	
	Type of specimens No (%)	Blood		24 (20)	
		Pus		23 (19.2)	
		Stool		5 (4.2)	
		Urine		53 (44.2)	
		Sputum		12 (10)	
		Endotracheal tube		1 (0.8)	
		Vaginal swab		1 (0.8)	
		Peritoneal fluid		1 (0.8)	

Table 2. Frequency of resistant genes in carbapenem-resistant *Enterobacteriaceae*.

Carbapenem-resistant *Enterobacteriaceae* (n = 26)	Resistant gene(s)					
	Triple	Double			Single	Total
	KPC/ NDM-1/ OXA-48	NDM1/ OXA-48	KPC/ OXA-48	Total	OXA-48	
K. pneumoniae	17 (65.4) [a]	2 (7.7)	2 (7.7)	4 (15.4)	0 (0)	21 (80.8) [b]
E. cloacae	0 (0)	1 (3.8)	0 (0)	1 (3.8)	1 (3.8)	2 (7.7)
E. coli	0 (0)	1 (3.8)	0 (0)	1 (3.8)	1 (3.8)	2 (7.7)
P. mirabilis	0 (0)	1 (3.8)	0 (0)	1 (3.8)	0 (0)	1 (3.8)
Total	17 (65.4) [c]	5 (19.2)	2 (7.7)	7 (26.9)	2 (7.7)	26 (100)

[a] Significantly higher frequency ($P \leq 0.001$) of triple gene resistance in *K. pneumoniae* isolates as compared to other types of isolates; [b] Significantly higher frequency ($P \leq 0.001$) of overall gene resistance in *K. pneumoniae* isolates as compared to other types of isolates; [c] Significantly higher frequency ($P = 0.001$) of triple gene resistance among all isolates as compared to double and single gene resistance.

recovered only from Saudi patients. In relation to gender, CRE isolates were more predominant in males (65%) compared to females (34.6%). Similarly, carbapenem-resistant *K. pneumoniae* isolates were also more predominant in males (50%) compared to females (30.8%). With regard to wards, the majority of CRE isolates were from ICU (53.9%), followed by medical (26.9%), surgical (11.5%) and pediatric (7.7%) wards. It was also revealed that 46.2% of resistant isolates recovered from ICU were *K. pneumoniae* compared to 23.1% from medical ward. Regarding to the type of specimens, a substantial number of CRE strains were isolated from blood (34.6%), followed by urine (23.1%), pus (19.2%) and sputum (15.5%) specimens. The only bacterium isolated from blood specimens was *K. pneumoniae*, which was also predominantly recovered from urine (19.2%) and sputum (15.5%).

Carbapenem resistance-associated genes

Molecular analysis of all 26 CRE isolates for carbapenemase genes revealed the presence of three known respective genes $bla_{\text{NDM-1}}$ (Figure 1), bla_{KPC} (Figure 2) and $bla_{\text{OXA-48}}$ (Figure 3). All 26 isolates were found positive for OXA-48, while the majority (24/26) had combination of carbapenemase genes. Significantly higher frequency ($P < 0.000$) of overall gene resistance was seen in *K. pneumoniae* isolates as compared to the other enterobacterial isolates. Out of 21 carbapenem-resistant *K. pneumoniae* isolates, 17 had triple gene resistance (KPC/NDM-1/OXA-48), and 4 had double gene resistance (2 isolates KPC/OXA-48 and 2 isolates

Figure 1. Representative 1% agarose gel showing positive and negative results of NDM resistance based on the amplification of corresponding $bla_{\text{NDM-1}}$ gene from suspected samples. L, 100 bp Ladder; lanes 2,4,5,6,7: representative positive samples showing specific 758 bp product; lanes 1, 3: representative negative strains showing no product and lane 8: negative control.

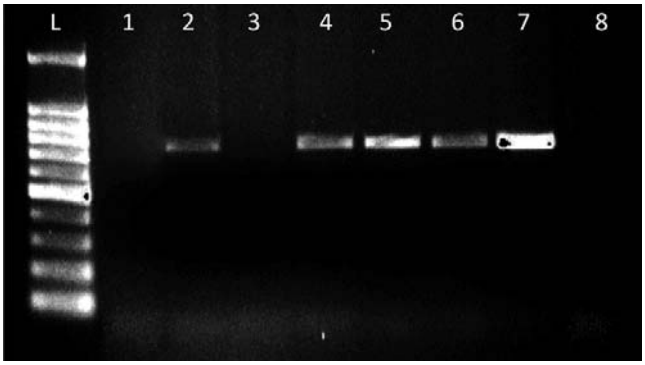


Figure 2. Representative 1% agarose gel showing positive and negative results of KPC resistance of all investigated isolates based on the amplification of corresponding bla_{KPC} gene. L, 100 bp Ladder; lanes 1, 2,4,5,7: representative positive strains showing specific 862 bp product; lanes 3,6: representative negative strains showing no product and lane 8: negative control.

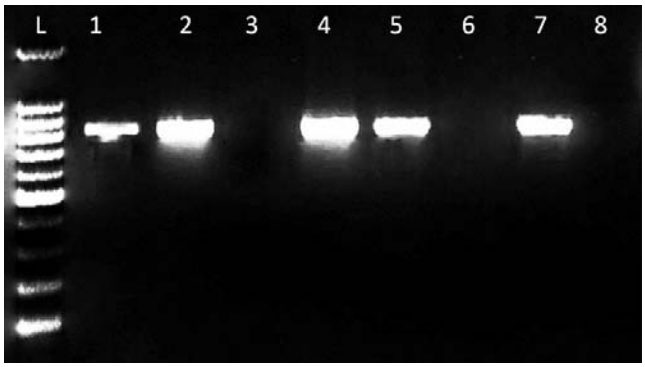
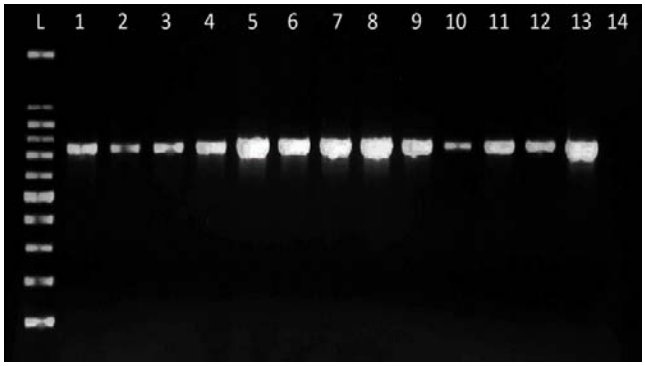


Figure 3. Representative 1% agarose gel showing positive results of OXA-48 resistance of all investigated isolates based on the amplification of corresponding $bla_{\text{OXA-48}}$ gene. L, 100 bp Ladder; lanes 1-13: representative positive strains showing specific 743 bp product and lane 14: negative control.



NDM-1/OXA-48). With regard to the other enterobacterial species, out of 2 isolates of both *E. coli* and *E. cloacae*, one isolate of each organism had combined resistant genes (NDM-1/ OXA-48) while the other isolates of each organism had only OXA-48 gene. In addition, single isolate of *P. mirabilis* carried the combined genes (NDM-1/OXA-48) (Table 2). Among all resistant isolates, triple gene resistance showed high significant frequency (P = 0.001) compared to double or single gene resistance. Moreover, triple gene resistance was the highest among *K. pneumoniae* isolates (P < 0.001) as compared to other enterobacterial isolates. (Table 2).

Triple gene resistance in K. pneumoniae isolates

The frequency and association risk of triple genes resistance in *K. pneumoniae* isolates in relation to nationality and gender of patients as well as type and source of specimens are shown in Table 3. With regards to gender, significant higher frequency (P = 0.028) was seen among male as compared to female patients with a significant association risk (COR 4.5; CI: 1.9-17.3; *P* = 0.018). Similarly, significant higher frequency (P = 0.0001) of triple gene resistance was evident among non-Saudi as compared to Saudi patients with a significant association risk (COR 4.9; CI: 1.5-19.3; *P* = 0.003). On the other hand, triple gene resistance showed significant higher frequency (P = 0.004) in ICU-obtained specimens as compared to the other sources with a significant association risk (COR 3.6; CI: 1.5-8.4; *P* = 0.002). With regard to specimen type, triple gene resistance showed significant higher frequency (P = 0.001 and P = 0.04) in blood specimen as compared to pus/sputum and urine, respectively with a significant association risk (COR 2.8; CI: 1.1-6.9; *P* = 0.02). No significant association pattern was evident between patients hospitalized in the same hospital/ward (Table 4-S).

Discussion

The emerging trends in global distribution of carbapenem resistant strains have intensified the need for regular surveillance of CRE in the region of Arabian Peninsula. The current study investigated the distribution and possible emergence of carbapenemases in *Enterobacteriaceae* isolates and its possible associated risk factors in Makkah, Western region, Saudi Arabia. The current study revealed *K. pneumoniae* as the most predominant (80.8%) resistant organism, which was almost equally distributed between Saudi and non-Saudi patients. Resistance in *K. pneumoniae* represents a considerable problem in the healthcare [1] and is mostly due to production of KPC that more frequently encountered in United States, Greece, South America and China [2]. In Saudi Arabia, although carbapenem resistance remained uncommon among *Enterobacteriaceae*, the first outbreak of carbapenem-resistant *K. pneumoniae* was reported in 2010 [30]. Later, the molecular basis of this resistance was found to be due to the involvement of OXA-48 in combination with CTX-M-15 genes [31]. Further studies detected OXA-48 and NDM-1 in carbapenem resistant *K. pneumoniae* isolates from Riyadh, Central region of Saudi Arabia [10,16]. However, isolated

Table 3. Incidence of triple genes among *K. pneumoniae* in relation to patient gender, specimen type and source.

Variables		Incidence of Triple genes No. (%)		
		Saudi (n = 27)	**Non-Saudi (n = 15)**	**Total (n = 42)**
Gender	Male	5 (18.5)	7 (46.6)	12 (28.6) [a]
	Female	1 (3.7)	4 (26.7)	5 (11.9)
	Total	6 (22.2)	11 (73.3) [b]	17 (40.5)
Specimen source	Intensive care unit	2 (7.4)	7 (46.7) [c]	9 (21.4) [d]
	Medical ward	3 (11.1)	2 (13.3)	5 (11.9)
	Surgical ward	0	2 (13.3)	2 (4.8)
	Pediatric ward	1 (3.7)	0	1 (2.4)
Specimen type	Blood	2 (7.4)	5 (33.3) [e]	7 (16.7) [f,g]
	Pus	0	2 (13.3)	2 (4.8)
	Urine	3 (11.1)	2 (13.3)	5 (11.9)
	Sputum	1 (3.7)	2 (13.3)	3 (7.1)

[a] Significant higher frequency (P = 0.028) among male as compared to female patients with significant association risk (COR 4.5; CI:1.97-17.265; P = 0.018); [b] Significant higher frequency (P = 0.0001) among non-Saudi as compared to Saudi patients with significant association risk (COR 4.9; CI: 1.5-19.3; P = 0.003); [c] Significant higher frequency (P < 0.0000) in ICU obtained specimens as compared to other sources among non-Saudi patients; [d] Significant higher frequency (P = 0.004) in ICU obtained specimens as compared to other sources among all patients with significant association risk (COR 3.6; 1.5-8.4; *P* = 0.002); [e] Significant higher frequency (P = 0.001) in blood specimens as compared to other specimen types among non-Saudi patients; [f,g] Significant higher frequency (P = 0.001) in blood specimen as compared to pus and sputum and (P = 0.04) as compared to urine, respectively with significant association risk (COR 2.8; CI: 1.1-6.9; (*P* = 0.02) in all patients.

strains neither harbored KPC resistant gene nor a combination of OXA-48 and NDM-1 genes [16]. The current study is the first to characterize the molecular basis of carbapenem resistance in Makkah, Western region of Saudi Arabia. In addition to reporting NDM and OXA-48-producing *Enterobacteriaceae* isolates, the current study also reported KPC-producing *K. pneumoniae* for the first time in Saudi Arabia. Moreover, this interesting finding represents the second record of KPC-producing *K. pneumoniae* in the Arabian Peninsula next to UAE [20].

Although rare, *K. pneumoniae* isolates producing combination of carbapenemase genes, such as NDM-1 and OXA-48 or NDM-1 and OXA-181 have been reported from Singapore and European and Middle East countries [15,22-25]. To the best of our knowledge, no previous studies have reported the co-existence of the triple carbapenemase genes in *K. pneumoniae* isolates. Remarkably, the current study not only reported the presence of double genes, NDM-1/OXA-48, in *Enterobacteriaceae* isolates, but also proved the presence of the 3 known carbapenem-resistant genes (KPC/NDM/OXA-48) in *K. pneumoniae* isolates from Makkah region for the first time.

Presence of the triple resistance genes, as revealed in the current study, was most predominant with a significant high frequency (P = 0.001) as compared to either double or single gene resistance among all CRE isolates. Interestingly, all cases of triple resistance genes were only reported in *K. pneumoniae* isolates (Table 2) with a significantly higher frequency (P = 0.0001) among non-Saudi patients (73.3%) as compared to Saudi patients (22.2%) (Table 3). One possible explanation of these findings could be attributed to the setting of the current study, which were conducted for the first time in Makkah region. In other words, the emergence of these resistant strains could be linked to the exceptional diversity of the population of the study setting. Makkah is a unique place in Saudi Arabia and the entire world. It annually receives more than three million pilgrims during the pilgrimage season, in addition to several other millions of visitors during the whole year [32,33]. The fact that many of those visitors and foreign residents come from endemic carbapenem resistant or high-incidence countries as India, Pakistan, Far East and Middle East [2,5,6,21] extremely increase the risk for emerging of new multidrug resistant strains from all over the world.

The antibiotic susceptibility profile of our isolates is similar to those reported from multidrug-resistant organisms harboring carbapenemase genes from the countries of the Middle East e.g., Kuwait [8], Lebanon [9], Saudi Arabia [16], UAE [34] and also other countries; e.g., USA [35] and China [36]. Some of the KPC-producing *K. pneumoniae* strains remain susceptible to gentamicin, but this is not the case for NDM- producing *Enterobacteriaceae* strains as many of which produce 16S ribosomal RNA methyltransferase that makes them highly resistant to all aminoglycosides including gentamicin. Though OXA-48 itself does not hydrolyze cephalosporins efficiently, OXA-48-producing *K. pneumoniae* strains appear to co-produce ESBL in most instances, therefore, they are resistant to cephalosporins as well as carbapenems [19]. This phenomenon has been observed by many investigators where multi-drug resistant isolates are often associated with carbapenemase production [15,35-36].

In our study, frequency and risk association to the triple resistance genes in *K. pneumoniae* isolates were significantly evident among overall male patients (COR 4.5; CI: 1.9-17.3; *P* = 0.018) as compared to female patients and was also evident among non-Saudi (COR 4.9; CI: 1.5-19.3; *P* = 0.003) as compared to Saudi patients. This could be linked to the population travelling to Saudi Arabia either for the employment or Islamic rituals in particular to Makkah throughout the year from countries known as common reservoirs for carbapenemase producing (NDM, OXA-48 and KPC) *Enterobacteriaceae* [6,13]. These factors greatly increase the risk of emerging new multidrug resistant strains, which make Makkah highly exposed to the spread of various infectious agents including CRE. On the other hand, the current study also revealed that the majority of the *K. pneumoniae* isolates carrying triple genes were isolated from blood specimens with a significant risk association (COR 2.8; CI: 1.1-6.9; *P* = 0.02) as compared to the other types of specimens collected from ICU patients (COR 3.6; CI: 1.5-8.4; *P* = 0.002) and other wards. These findings draw the attention towards nosocomial infections as a possible source of dissemination of these resistance strains among critically ill patients. Similar findings have been reported in a study from Riyadh, Saudi Arabia, where *K. pneumoniae* harboring NDM genes were isolated from ICU patients [16].

At present, it is difficult to predict whether the occurrence of these carbapenemases-producing *K. pneumoniae* isolates is due to population movement or local emergence of these strains, in particular KPC-positive *K. pneumoniae*, which has not been reported earlier from Saudi Arabia. Interestingly, previous studies from Arabian Peninsula reported that CRE infections are not limited to foreign exposure. It was

shown that the majority of reported cases had no history of foreign travel or hospitalization abroad [7,8,10,15]. In UAE, a recent study showed that lack of foreign exposure was more common in OXA-48-like carbapenemase producing and VIM positive strains where the majority of these strains appeared to be acquired locally. On the other hand, previous hospitalization abroad (mostly in India) could represent a risk factor for acquisition of NDM-1 gene [35]. Thus, the currently isolated triple genes-producing *K. pneumoniae* may suggest that in addition to international travel exposure, indigenous transmission could be a major factor for the emergence of those strains as well.

In the current study, certain limitations were evident including the lack of international travel and/or foreign hospitalization history of the investigated patients. Another limitation is the lack of efficient admission screening and information about the history of patients whether they were carrying carbapenemase-producing pathogen at the time of admission or were nosocomial acquisition. Indeed, further epidemiologic and genotypic investigations are needed to be conducted to unveil the curiosity behind the multiple genes-based resistance and to trace the source of these emerging extraordinary resistant isolates.

Conclusion

This study documents the ongoing emergence of carbapenemase genes (KPC, NDM-1 and OXA-48) and in particular the coexistence of these genes in *K. pneumoniae* strains. These resistant isolates showed significant risk association with the male gender (COR 4.5; CI: 1.9-17.3; $P = 0.018$) and non-Saudi nationality (COR 4.9; CI: 1.5-19.3; $P = 0.003$) as well as blood specimens (COR 2.8; CI: 1.1-6.9; $P = 0.02$) of ICU patients (COR 3.6; CI: 1.5-8.4; $P = 0.002$). Such isolates can have significant public health importance as these may combine a variety of resistance attributes that make them nearly untreatable. Therefore, there is a need to extend the awareness among healthcare providers in the countries. Additionally, improved national surveillance mechanisms for the detection of these multidrug-resistant pathogens will help to take appropriate infection control measures in order to curtail further dissemination of these life-threatening pathogens. At the same time, close cooperation between the countries of the region is required to control the dissemination across borders.

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Annex – Supplementary Items

Supplementary Table 1. Antibiotic susceptibility profile of isolates investigated for carbapenemase genes in relation to corresponding patient, specimen and isolates data

Isolate No.	Organism	Ward	Specimen	Nationality	Carbapenemase genes	Ceftazidime	Cefotaxime	Ceftriaxone	Cefepime	Imipenem	Meropenem	Amikacin	Gentamicin	Colistin
C-1	*K. pneumoniae*	ICU	Sputum	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
C-2	*K. pneumoniae*	Pediatric ward	Blood	Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
C-3	*K. pneumoniae*	Medical ward	Blood	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
C-4	*K. pneumoniae*	ICU	Blood	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
C-5	*K. pneumoniae*	ICU	Blood	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
C-6	*K. pneumoniae*	ICU	Blood	Saudi	NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
C-7	*K. pneumoniae*	ICU	Blood	Saudi	KPC, OXA-48	R	R	R	R	R	R	S	S	S
C-8	*E. cloacae*	ICU	Pus	Saudi	NDM-1, OXA-48	R	R	R	R	R	R	R	S	S
C-9	*Escherichia coli*	Medical ward	Stool	Saudi	NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
C-10	*K. pneumoniae*	ICU	Sputum	Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-11	*K. pneumoniae*	ICU	Sputum	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-12	*K. pneumoniae*	ICU	Sputum	Saudi	NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-13	*K. pneumoniae*	Medical ward	Urine	Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-14	*K. pneumoniae*	Surgical ward	Pus	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-15	*K. pneumoniae*	ICU	Urine	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-16	*K. pneumoniae*	Medical ward	Urine	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-17	*K. pneumoniae*	ICU	Blood	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-18	*K. pneumoniae*	ICU	Blood	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
C-19	*K. pneumoniae*	Medical ward	Urine	Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
C-20	*K. pneumoniae*	ICU	Blood	Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
C-21	*Escherichia coli*	Pediatric ward	Urine	Saudi	OXA-48	R	R	R	R	R	R	S	R	S
C-22	*K. pneumoniae*	Surgical ward	Pus	Non-Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
C-23	*K. pneumoniae*	Medical ward	Endo tracheal tube	Saudi	KPC, OXA-48	R	R	R	R	R	R	R	R	S
C-24	*P. mirabilis*	Surgical ward	Pus	Saudi	NDM-1, OXA-48	R	R	R	R	R	R	S	R	R
C-25	*K. pneumoniae*	Medical ward	Urine	Saudi	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
C-26	***E. cloacae***	**ICU**	**Pus**	**Saudi**	**OXA-48**	**R**	**R**	**R**	**R**	**R**	**R**	**S**	**I**	**S**

KPC, *Klebsiella pneumoniae* carbapenemase; NDM-1, New Delhi metallo-ß-lactamase-1; OXA-48, oxacillinase-48; ICU, intensive care unit; R, resistant; S, sensitive.

Supplementary Table 2. Minimum inhibitory concentration of isolates carrying NDM-1, KPC and OXA-48 genes.

No.	Organism	Ceftazidime	Cefotaxime	Ceftriaxone	Cefepime	Imipenem	Meropenem	Amikacin	Gentamicin	Colistin
C-1	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	4 (S)	2 (S)
C-2	*K.pneumoniae*	> 16 (R)	> 32 (R)	> 64 (R)	> 32 (R)	> 8 (R)	> 8 (R)	< 16 (S)	> 8 (R)	< 0.5 (S)
C-3	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	< 1 (S)	< 0.5 (S)
C-4	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	< 1 (S)	< 0.5 (S)
C-5	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	< 1 (S)	2 (S)
C-6	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	< 1 (S)	< 0.5 (S)
C-7	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	< 1 (S)	< 0.5 (S)
C-8	*E.cloacae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 4 (R)	> 64 (R)	< 1 (S)	< 0.5 (S)
C-9	*Escherichia coli*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	< 2 (S)	> 16 (R)	2 (S)
C-10	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	8 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-11	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	8 (R)	> 16 (R)	> 64 (R)	> 16 (R)	2 (S)
C-12	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-13	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-14	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	4 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-15	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	> 64 (R)	> 16 (R)	2 (S)
C-16	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	8 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-17	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-18	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	> 64 (R)	> 16 (R)	< 0.5 (S)
C-19	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	8 (R)	> 16 (R)	8 (S)	> 16 (R)	2 (S)
C-20	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	4 (S)	> 16 (R)	< 0.5 (S)
C-21	*Escherichia coli*	> 16 (R)	> 32 (R)	> 64 (R)	> 32 (R)	> 8 (R)	> 8 (R)	< 16 (S)	> 16 (R)	< 0.5 (S)
C-22	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 32 (R)	> 16 (R)	> 16 (R)	16 (S)	2 (S)	< 0.5 (S)
C-23	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	64 (R)	> 16 (R)	2 (S)
C-24	*P.mirabilis*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	< 2 (S)	> 16 (R)	> 16 (R)
C-25	*K.pneumoniae*	> 64 (R)	> 32 (R)	> 64 (R)	> 64 (R)	> 16 (R)	> 16 (R)	16 (S)	> 16 (R)	< 0.5 (S)
C-26	*E.cloacae*	> 64 (R)	> 32 (R)	> 64 (R)	32 (R)	> 16 (R)	> 16 (R)	16 (S)	8 (I)	< 0.5 (S)

KPC, *Klebsiella pneumoniae* carbapenemase; NDM-1, New Delhi metallo-ß-lactamase-1; OXA-48, oxacillinase-48; R, resistant; S, sensitive.

Supplementary Table 3. Antibiotic susceptibility profile of all investigated *Enterobacteriaceae* strains from participating hospitals (n = 120).

S. No	Hospital code No **	Organism	Carbapenemase genes	Antibiotics								
				Ceftazidime	Cefotaxime	Ceftriaxone	Cefepime	Imipenem	Meropenem	Amikacin	Gentamicin	Colistin
1	B-558 (C4)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
2	B-590 (C5)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
3	B-1036 (C6)	*K. pneumoniae*	NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
4	B-43	*E. cloacae*	---	R	R	R	R	S	S	S	R	S
5	B-880	*Escherichia coli*	---	I	R	R	I	S	S	S	R	S
6	B-44	*E. cloacae*	---	R	R	R	R	S	S	R	S	S
7	B-1004	*Escherichia coli*	---	S	R	I	S	S	S	S	S	S
8	B-1041(C7)	*K. pneumoniae*	KPC, OXA-48	R	R	R	R	R	R	S	S	S
9	B-5048 (C26)	*E. cloacae*	OXA-48	R	R	R	R	R	R	S	I	S
10	B-5056	*K. pneumoniae*	---	R	R	R	R	S	S	S	R	S
11	B-28 (C3)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
12	B-443	*Escherichia coli*	---	R	R	R	R	S	S	S	R	S
13	B-5177	*K. pneumoniae*	---	R	R	R	R	S	S	S	S	S
14	B-5230	*P. mirabilis*	---	R	R	R	R	S	S	R	S	R
15	B-5228	*Escherichia coli*	---	S	R	R	R	S	S	S	R	S
16	B-5229	*Escherichia coli*	---	S	S	S	S	S	S	S	R	S
17	B-5259	*K. pneumoniae*	---	I	R	R	I	S	S	S	S	S
18	B-5230 (C8)	*E. cloacae*	NDM-1, OXA-48	R	R	R	R	R	R	R	S	S
19	B-1068	*Escherichia coli*	---	R	R	R	R	S	S	S	R	S
20	B-1322	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
21	B-5100	*K. ornithinolytica*	---	R	R	I	S	s	s	S	R	S
22	K-1 (C9)	*Escherichia coli*	NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
23	K-2	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
24	Z-1	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
25	Z-2G(C24)	*Proteus mirabilis*	NDM-1, OXA-48	R	R	R	R	R	R	S	R	R
26	Z-2Y	*K. pneumoniae*	---	R	R	R	R	S	S	R	R	S
27	Z-3	*Escherichia coli*	---	R	R	R	R	S	S	S	I	S
28	Z-4(C25)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
29	Z-6	*E. coli*	---	S	S	S	S	S	S	S	S	S
30	Z-8	*E. coli*	---	R	I	S	S	S	S	S	S	S
31	Z-9	*E. coli*	---	S	S	S	S	S	S	S	S	S
32	Z-10	*E. coli*	---	S	S	S	S	S	S	S	S	S
33	Z-11Y	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
34	Z-11G	*Proteus mirabilis*	---	S	S	S	S	S	S	S	R	R
35	Z-12	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
36	Z-13(C1)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
37	Z-17	*Citrobacter*	---	S	I	R	S	S	S	R	S	S
38	Z-18	*K. ornithinolytica*	---	S	R	R	S	S	S	S	S	S
39	Z-19	*K. ornithinolytica*	---	S	S	S	S	S	S	S	S	S
40	Z-21	*E. coli*	---	S	R	R	S	S	S	S	S	S
41	Z-23	*Proteus mirabilis*	---	R	R	I	S	S	S	S	R	R
42	Z-25	*Citrobacter*	---	I	R	R	I	S	S	S	S	S
43	Z-24	*Proteus mirabilis*	---	S	R	R	I	S	S	S	S	R
44	Z-26	*K. pneumoniae*	---	S	S	S	S	S	S	S	R	S
45	Z-27(C22)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	S	S
46	Z-28	*Proteus mirabilis*	---	S	S	S	S	S	S	S	R	R
47	Z-29	*K. pneumoniae*	---	I	R	R	I	S	S	S	S	S
48	F-1	*E.coli*	---	R	R	R	R	S	S	S	S	S
49	F-2 (C10)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
50	F-3	*E. coli*	---	S	S	S	S	S	S	S	R	S
51	F-4	*Serratia sp.*	---	R	R	R	R	S	S	I	R	S

S. No	Hospital code No **	Organism	Carbapenemase genes	Antibiotics								
				Ceftazidime	Cefotaxime	Ceftriaxone	Cefepime	Imipenem	Meropenem	Amikacin	Gentamicin	Colistin
52	F-5	*Serratia sp.*	---	R	R	R	R	S	S	S	S	S
53	F-7	*Serratia sp.*	---	S	S	S	S	S	S	S	S	S
54	F-8	*Serratia sp.*	---	R	R	R	R	S	S	S	R	S
55	F-9	*K. oxytoca*	---	I	R	R	I	S	S	S	S	S
56	F-10	*Serratia sp.*	---	S	S	S	S	S	S	S	S	S
57	F-11 (C11)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
58	F-12 (C12)	*K. pneumoniae*	NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
59	F-13	*R. ornithinolytica*	---	R	R	R	R	S	S	S	S	S
60	F-14	*Citrobacter*	---	S	R	R	S	S	S	S	R	S
61	F-15	*K. pneumoniae*	---	R	R	R	R	S	S	S	S	S
62	F-16(C13)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
63	F-17(C14)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
64	F-18	*K. oxytoca*	---	S	R	I	S	S	S	S	S	S
65	F-19	*K. oxytoca*	---	R	R	R	R	S	S	S	R	S
66	F-20(C15)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
67	F-21	*K. oxytoca*	---	R	R	R	R	S	S	S	R	S
68	F-22	*S. liquefaciens*	---	R	R	S	R	S	S	S	S	S
69	F-23	*E. coli*	---	R	R	R	R	S	S	S	S	S
70	F-24	*E. coli*	---	I	R	R	I	S	S	S	R	S
71	F-26(C16)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
72	F-27(C17)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
73	F-28	*K. pneumoniae*	---	I	R	R	S	S	S	S	S	S
74	F-29	*E. coli*	---	S	S	S	S	S	S	S	S	S
75	F-30	*E. coli*	---	S	S	S	S	S	S	S	S	S
76	F-31(C18)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	R	R	S
77	F-32	*E. coli*	---	S	R	R	S	S	S	S	R	S
78	F-33	*E. coli*	---	S	R	R	S	S	S	S	S	S
79	F-34(C19)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
80	F-35	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
81	F-36	*Citrobacter*	---	S	R	R	S	S	S	S	R	S
82	F-37	*K. pneumoniae*	---	R	R	R	R	S	S	S	R	S
83	F-40	*K. pneumoniae*	---	S	S	S	S	S	S	S	S	S
84	F-41	*K. pneumoniae*	---	R	R	R	R	S	S	S	S	S
85	F-42(C20)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
86	F-43	*E. coli*	---	R	R	R	R	S	S	S	S	S
87	MCH-1275(C2)	*K. pneumoniae*	KPC, NDM-1, OXA-48	R	R	R	R	R	R	S	R	S
88	MCH-1299	*Escherichia coli*	---	R	R	R	R	S	S	S	S	S
89	MCH-1288	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
90	MCH-664BC	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
91	MCH-1241	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
92	MCH-1264	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
93	MCH-109	*K. oxytoca*	---	R	S	I	R	S	S	S	S	S
94	MCH-123	*Escherichia coli*	---	R	R	R	I	S	S	S	R	S
95	MCH-1318	*Salmonella*	---	S	S	S	S	S	S	S	S	S
96	MCH-272	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
97	MCH-279	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
98	MCH-429	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
99	MCH-440	*Escherichia coli*	---	S	S	S	S	S	S	S	S	S
100	MCH-455	*Escherichia coli*	---	S	S	R	S	S	S	S	R	S
101	MCH-378	*Escherichia coli*	---	S	S	R	R	S	S	S	R	S
102	MCH-482	*Escherichia coli*	---	S	S	S	S	S	S	S	R	S
103	MCH-494	*Escherichia coli*	---	S	S	R	S	S	S	S	S	S

S. No	Hospital code No **	Organism	Carbapenemase genes	Antibiotics								
				Ceftazidime	Cefotaxime	Ceftriaxone	Cefepime	Imipenem	Meropenem	Amikacin	Gentamicin	Colistin
104	MCH-723(C21)	*Escherichia coli*	OXA-48	R	R	R	R	R	R	S	R	S
105	MCH-163 (C23)	*K. pneumoniae*	KPC, OXA-48	R	R	R	R	R	R	R	R	S
106	G-1	*Salmonella sp.*	----	S	S	S	S	S	S	S	S	S
107	G-2	*E. coli*	----	S	R	S	R	S	S	S	R	S
108	G-4	*E. aerogenes*	----	S	S	S	S	S	S	S	R	S
109	G-5	*S. liquefaciens*	----	R	R	R	S	S	S	S	S	S
110	G-6	*E. coli*	----	S	S	S	S	S	S	S	R	S
111	G-7	*K. ornithinolytica*	----	S	S	S	S	S	S	S	S	S
112	G-8	*K. pneumoniae*	----	S	S	S	S	S	S	S	S	S
113	G-10	*E. coli*	----	S	R	R	I	S	S	S	R	S
114	G-11	*K. ornithinolytica*	----	S	S	S	S	S	S	S	R	S
115	G-12	*E. cloacae*	----	S	S	S	S	S	S	S	S	S
116	G-13	*K. pneumoniae*	----	R	R	R	I	S	S	S	S	S
117	G-14	*K. pneumoniae*	----	R	R	R	S	S	S	S	R	S
118	G-15	*K. pneumoniae*	----	R	R	R	R	S	S	S	S	S
119	G-17	*Salmonella sp.*	----	S	S	S	S	S	S	S	R	S
120	G-18	*E. coli*	----	S	S	S	S	S	S	S	R	S

** Hospital code key: B & K = Al-Noor Specialized hospital (n = 23); Z = King Abdul Aziz Hospital (n = 24); F = King Faisal Hospital (n = 39); MCH & G = Maternity and Children Hospital (n = 34)

Supplementary Table 4. Distribution of *Klebsiella penumoniae* isolates carrying triple genes in relation to hospitals/wards.

Hospitals	***Klebsiella penumoniae* isolates with triple genes**				
	Intensive care unit	Medical ward	Surgical ward	Pediatric ward	Total
King Faisal hospital	5	2	2	1	10
Maternity and children hospital	0	1	0	0	1
Al-Noor specialist hospital	2	1	0	0	3
King Abdul Aziz hospital	2	1	0	0	3
Total	9	5	2	1	17