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Assessment of Carbapenem-Resistant *Enterobacteriaceae* and Other Gram-Negative Bacteria on Intensive Care Unit Surfaces and Among Healthcare Workers at Tobruk Medical Center

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ABSTRACT

The emergence of carbapenem-resistant Enterobacteriaceae (CRE) and other multidrug-resistant Gram-negative bacteria (GNB) presents a serious threat to infection control, particularly in high-risk environments such as Intensive Care Units (ICUs). ICU surfaces and healthcare workers (HCWs) may serve as reservoirs and transmission routes for these pathogens. This study aimed to assess the prevalence and distribution of CRE, and other Gram-negative bacteria isolated from ICU environmental surfaces and from the nasal swab of healthcare workers. A descriptive cross-sectional study was used to study 127 collected samples: 36 from healthcare workers and 91 from surface smears. Samples were collected from four units of intensive care: Intensive Care Unit (ICU), Cardiac Care Unit (CCU), Pediatric Care Unit (PICU), and Neonatal Intensive Care Unit (NICU). Identified Enterobacteriaceae were confirmed by biochemical methods, and tested for carbapenemase production using three discs (EME, IMP, ETP). Resistance to Ertapenem, Imipenem, and Meropenem was considered carbapenem resistance. A total of 13 (36.1%) out of 36 HCW samples yielded clinical isolates: 3 *Pantoea* sp., 3 *Klebsiella pneumoniae*, 2 *Acinetobacter baumannii*, 2 *Escherichia coli*, 2 *Flavimonas oriyzihabitans*, and 1 *Serratia marcescens*. One (7.69%) isolate showed carbapenem resistance: 1 *Klebsiella pneumonia*. From 91 surface samples, 39 (43%) yielded isolates: 13 *Flavimonas oriyzihabitans*, 6 *Acinetobacter baumannii*, 5 *Pantoea* sp., 4 *Escherichia coli*, 3 *Enterobacter cloacae*, 2 *Serratia marcescens*, 2 *Shigella* sp., 1 *Klebsiella pneumonia*, 1 *Leclercia adecarboxylata*, and 1 *Salmonella* sp. Of these, 11 (28.2%) were carbapenem-resistant: 3 *Escherichia coli*, 3 *Acinetobacter baumannii*, 2 *Pantoea* sp., 1 *Klebsiella pneumonia*, 1 *Citrobacter freundii*, and 1 *Enterobacter cloacae*.

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INTRODUCTION

Hospital-acquired infections (HAIs) are a serious worldwide concern, particularly in critical care units (ICUs), where patients are more vulnerable because of immunosuppression, invasive procedures, and extended hospital stays. A major public health worry nowadays is the emergence of multidrug-resistant Gram-negative bacteria (GNB), including carbapenem-resistant *Enterobacteriaceae* (CRE) [1]. The pathogens that cause HAIs are gram-negative bacteria (GNB), especially *Enterobacteriaceae*. They have shown that antimicrobial resistance is on the rise, including resistance to medications like carbapenems that are employed as last resorts [2, 3].

Carbapenem-resistant *Enterobacteriaceae* (CRE), including Escherichia coli and *Klebsiella pneumoniae*, are especially troublesome because they can manufacture carbapenemases (e.g., KPC, NDM, OXA-48), which make most β-lactam antibiotics ineffective. CRE is a major priority pathogen group, according to the World Health Organization (WHO), necessitating immediate innovative treatment approaches and increased infection control measures [4]. High-touch surfaces like ventilators, bed rails, and monitors in the intensive care unit (ICU) can contain these resistant germs and operate as a continuous source of transmission. Furthermore, healthcare workers (HCWs) may act as vectors, promoting cross-contamination between patients and contaminated surfaces, if they do not carefully follow hand hygiene and personal protective equipment regulations [5, 6]. Studies in many developing nations still show a lack of use of environmental surveillance and resistance profiling, despite awareness of these risks. To enhance infection prevention and control measures, it is critical to assess the presence of CRE and other GNB in intensive care units and among healthcare workers. Multidrug-resistant GNB and CRE have alarmingly increased in hospital settings

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in Libya, according to recent research [7,8]. There is, however, a dearth of information explicitly addressing environmental contamination in intensive care units and the function of healthcare workers. The development of targeted infection is hampered by this knowledge gap. Gram-negative, rod-shaped, facultatively anaerobic, nonsporulating proteobacteria make up the diverse *Enterobacteriaceae* family. The majority of this family's members have Type 1 fimbriae, or pili, which are essential. They vitally facilitate bacterial movement. Some species can move and adhere to epithelial cells because they have flagella. The ability of Enterobacteriaceae to ferment a variety of sugars—some species focusing on specific sugars—is one of their key biochemical characteristics. These bacteria can grow in a variety of environments, such as water, soil, plants, and mammal gastrointestinal tracts, with ideal growth temperatures falling between 25°C and 37°C [9]. Infections acquired in the community and those caused by nosocomial infections are often attributed to the *Enterobacteriaceae*. They can gain genetic material through horizontal gene transfer, mostly through plasmids and transposons, and they can spread easily from person to person and through tainted food and water sources [10]. According to Kachrimanidou et al. [11], these microbes make up a sizable amount of the normal gut microbiota and are common in human illnesses. *Enterobacteriaceae* that are clinically relevant can be divided into two major groups: opportunistic pathogens like Enterobacter, Citrobacter, and Klebsiella, and primary pathogens like *Salmonella* and *Escherichia coli* [12].

METHODS

Site of study

This study was carried out at Tobruk Medical Centre, the city of Tobruk, Eastern Libya.

Sample collection

The samples or specimens were collected from healthcare workers (HCWs) / medical staff and surfaces of intensive care units. A total of 127 samples were collected from which 36 were taken from healthcare workers using nasal swabs and 91 smears were taken using cotton swabs from all surfaces and environment surrounding the patients, including medical devices, beds, tables, ventilators, nurseries within the departments of intensive care unit namely: the Intensive Care Unit (ICU), and Cardiac Care Unit (CCU), and Pediatric Intensive Care Unit (PICU), Neonatal Intensive Care Unit (NICU). The samples were taken over a period of 8 months from January 2024 to August 2024.

Inclusion criteria

This study on carbapenem-resistant Enterobacteriaceae at the Tobruk Medical Center was carried out and followed the ethical standards to protect participants. Ethical considerations include informed consent, voluntary participation, and confidentiality. The research protocol was reviewed before the samples and data collection. The risks and purpose of the study were explained to participants, and adhering to ethical considerations is important for the study's integrity and trust between researchers and participants.

Cultivation of samples

The collected samples/specimens were cultured on the blood agar to grow different types of bacteria of medical importance, such as Staphylococci and Streptococcus, gram-positive bacteria, and several types of gram-negative bacteria. Then the positive isolates were cultured on MacConkey agar; this medium is specifically designed for the growth of gram-negative bacteria, and were incubated at a temperature of 37°C for a period of 8-24 hours for the purpose of isolating them without color, and which were tested by the API-E20 (Analytical Profile Index) system and the biochemical tests.

Microbiological investigation

All samples collected from HCWs and surfaces of intensive care units were processed according to the procedure in Figure 1. Biochemical methods first confirmed the identified Enterobacteriaceae ,while the confirmed isolates were tested for carbapenemase production using three discs (EME , IMP ,and ETP .(The resistance to EME, IMP, and ETP was considered as CRE (Figure 1).

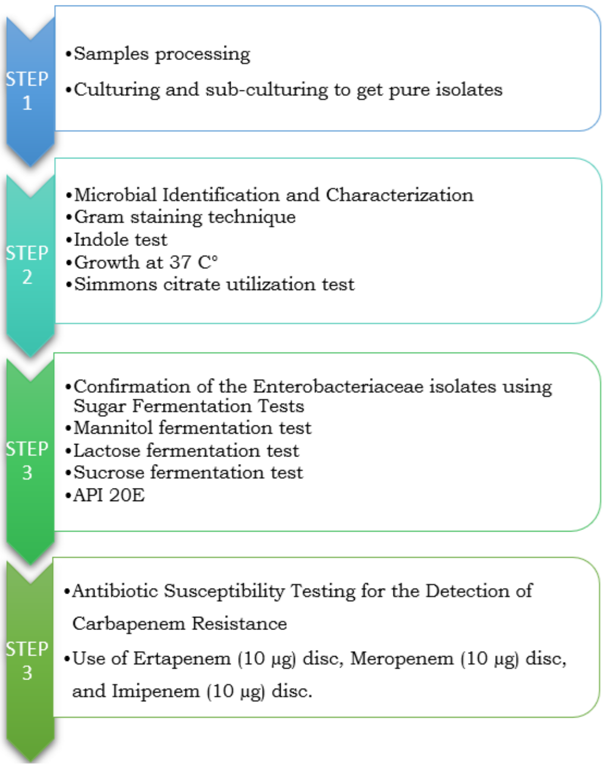

Figure 1: A flowchart of the detection of the Carbapenem-Resistant Enterobacteriaceae

Statistical analysis

Statistical analyses were done by using SPSS. Microsoft Office Excel 2019 was used for entry data. The results were expressed as percentages and proportions. The Chi-square test was performed. The Value of P≤0.05 was used as a significant level for association in comparison.

RESULTS

Description of study sample

A total of 127 samples were recruited and collected from Intensive Care Units (ICUs) at the Medical Center of Tobruk. From the above sample, 36 (28%) nasal swabs were collected from Health Care Workers (HCWs) who are working in these units, and 91 (72%) swab samples from different surfaces inside intensive care units (Figure 2).

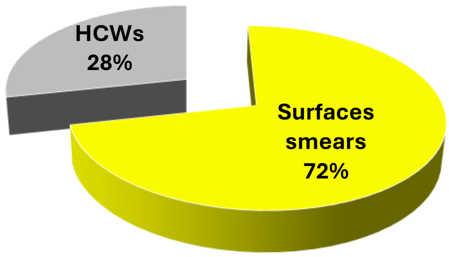

Figure 2: Description of study samples

Health care workers (HCWs) samples

In this study, 36 HCW members were investigated and included medical doctors, nurses, and other HCWs in the ICUs units. Only nine samples were collected from each intensive care unit: ICU, CCU, PICU, and NICU. However, 10 (27.8%) invested HCWs were male and 26 (72.2%) were females. All samples were collected by nasal swabs (Table 1).

Distribution of Gram-negative and Gram-positive bacteria among isolates from HCWs' samples

The results showed that the total percentage of Gram-negative bacteria isolated from health workers' samples from Tobruk Medical Center was 36.1%, while the total percentage of Gram-positive bacteria was 63.9% and relatively the highest (Table 2) (Figure 3). In the cardiac care unit (CCU), Gram-negative bacteria constituted 55.6% compared to 44.4% of Gram-positive bacteria, but the intensive care unit showed a slightly higher percentage of Gram-positive bacteria (66.7%) compared to Gram-negative bacteria (33.3%). On the other hand, the results showed that the percentage of Gram-negative bacteria in both the neonatal intensive care unit and the pediatric intensive care unit was (33.3%) and (22.2%), respectively. The highest percentage of Gram-positive bacteria was in the pediatric intensive care unit (77.8%), while in the neonatal intensive care unit, it was (66.7%). The result revealed that there was an insignificant (X2=2.28; P>0.05) association between intensive care unit and isolation of gram-positive and negative bacteria among HCWs (Table 3).

Table. 1: HCWs distribution by gender

Units	Source of specimen	No. (%) of isolate		
		Specimens No.	Gender	
			Male N(%)	Female N(%)
ICU	Nasal	9	2 (22.2)	7(77.8)
CCU	Nasal	9	4 (44.4)	5(55.6)
PICU	Nasal	9	3(33.3)	6 (66.7)
NICU	Nasal	9	1(11.1)	8 (88.9)
Total		36	10(27.8)	26(72.2)

Table 2: Distribution of culture results collected from HCWs

Units	Gram negative Bacteria N(%)	Gram positive Bacteria N(%)	Total N(%)
ICU	3(33.3)	6 (66.7)	9(100)
CCU	5(55.6)	4(44.4)	9(100)
PICU	2 (22.2)	7(77.8)	9(100)
NICU	3(33.3)	6(66.7)	9(100)
Total	13(36.1)	23(63.9)	36(100)
Chi-square test	X^2=2.28; P>0.05		

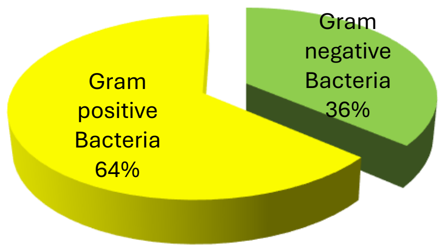

Figure 3: Distribution of Gram-negative and Gram-positive bacteria among isolates from HCWs

Of the 13 *Enterobacteriaceae* and gram-negative bacterial infections isolated from healthcare workers, the results showed that only 1 sample (7.69%) showed apparent resistance to carbapenem, and represented one bacterial species out of the six isolated. The *Klebsiella pneumoniae* (n = 1; 33.3%) showed carbapenem resistance (Table 4). The result of statistical analysis revealed an insignificant association (X2=3.61; P>0.05) between bacteria isolated from healthcare workers and carbapenem susceptibility.

Table 3: Distribution of organisms isolated from HCWs during the study period

Organism	**G⁻ isolated; n(%=n/ total isolate)**	**Carbapenem-susceptibility**	
		Sensitive; n (%=n/total organism)	Resistant; n (%=n/total organism)
Pantoea sp.	3(23.1)	3(100)	0(0)
Klebsiella pneumoniae	3(23.1)	2(66.7)	1(33.3)
Acinetobacter baumannii	2(15.4)	2(100)	0(0)
Flavimonas oriyzihabitans	2(15.4)	2(100)	0(0)
Escherichia coli	2(15.4)	2(100)	0(0)
Serratia marcescens	1(7.7)	1(20)	0(0)
Total	13(100)	12(92.30)	1(7.69)
Chi-square test	X^2=3.61; P>0.05		

Table 4: The antibiotic susceptibility pattern of isolated bacteria against carbapenems isolated from HCWs

Bacteria strain	**Number of CRE isolates**	**ERT**	**EME**	**IMP**
Klebsiella pneumoniae	1	R	R	R

Surfaces smears

Ninety-one smear samples were collected from the surfaces of ICU units and investigated. A total of 24 samples (26.4%) were collected from the ICU, 22 (24.2%) from the CCU, 22 (24.2%) from the PICU, and 23 (25.3%) from the NICU unit (Table 5).

Table 5: Surfaces smear distribution by the Medical Centre of Tobruk

Units	Number of samples	Percent (%)
ICU	24	(26.4)
CCU	22	(24.2)
PICU	22	(24.2)
NICU	23	(25.3)
Total	91	(100)

Distribution of Gram-negative and Gram-positive bacteria among isolates from surface smears

The results showed that the total percentage of Gram-negative bacteria isolated from surface smear samples from Tobruk Medical Center was 42.9%, while the total percentages of Gram-positive bacteria and no growth bacteria were 24.4% and 19.3%, respectively. In the ICU unit, Gram-negative bacteria constituted 37.5% compared to 12.5% of Gram-positive bacteria, but the CCU unit showed slightly higher percentages of Gram-positive bacteria (45.5%) and Gram-negative bacteria (45.5%). The percentage of Gram-negative bacteria in the PICU unit was the highest (54.5%), while the percentage of Gram-negative bacteria in the NICU unit was the lowest (34.8%). Also, the results showed that the highest percentage of no growth was recorded in the NICU, with 30.4%. The analysis of the chi-square test explained a significant (X2=16.15; P<0.05) association between intensive

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care unit and the isolated microorganism from surfaces (Table 6) (Figure 4).

Table 6: Distribution of culture results collected from surface smears

Units	**Gram-negative Bacteria N (%)**	**Gram-positive Bacteria N (%)**	**No growth N (%)**	**Total N (%)**
ICU	9(37.5)	3 (12.5)	12(50.0)	24(100)
CCU	10(45.5)	10(45.5)	2(9.1)	22(100)
PICU	12 (54.5)	8(36.4)	2(9.1)	22(100)
NICU	8(34.8)	8(34.8)	7(30.4)	23(100)
Total	39(42.9)	29(31.9)	23(25.2)	91(100)
Chi-square test	X^2=16.15; P<0.05			

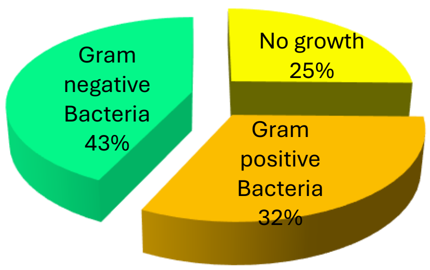

Figure 4: Distribution of Gram-negative and Gram-positive bacteria among isolates from surface smears

Of the 39 *Enterobacteriaceae* and gram-negative bacterial infections isolated from surface smears, 11(28.2%) displayed phenotypic carbapenem resistance, representing 6 different bacterial species of the 11 species studied. Of these, *Acinetobacter baumannii* (n = 3/39; 8%), *Escherichia coli* (n = 3/39; 8%), displayed the highest levels of carbapenem resistance. The other strains detected include: *Pantoea* sp. (n = 2/39; 5%), *Citrobacter freundii* (n = 1/39; 2.7%), *Klebsiella pneumoniae* (n = 1/39; 2.7%), and *Enterobacter cloacae* (n = 1/39; 2.7%). The result of chi-square explained a significant (X2=18.67; P<0.05) association between the isolated bacteria from surfaces and their carbapenem-susceptibility (Table 7).

Table 7: Distribution of organisms isolated from surface smears during the study period

Organism	G^- isolated; n(%=n/ total isolate)	Carbapenem-susceptibility	
		Sensitive; n (%=n/total organism)	Resistant; n (% =n/total organism)
Pantoea sp.	5(12.8)	3(60)	2(40)
Klebsiella pneumoniae	1(2.56)	0(0)	1(100)
Acinetobacter baumannii	6(15.4)	3(50)	3(50)
Flavimonas oriyzihabitans	13(33.3)	13(100)	0(0)
Escherichia coli	4(10.3)	1(25)	3(75)
Citrobacter freundii	1(2.56)	0(0)	1(100)
Enterobacter cloacae	3(7.7)	2(66.7)	1(33.3)
Serratia marcescens	2(5.1)	2(100)	0(0)
Leclercia adecarboxylat	1(2.56)	1(100)	0(0)
Salmonella spp.	1(2.56)	1(100)	0(0)
Shigella spp.	2(5.1)	2(100)	0(0)
Total	39(43)	28(71.8)	11(28.2)
Chi-square test	X^2=18.67; P<0.05		

Table 8: The antibiotic susceptibility pattern of isolated bacteria against carbapenems isolated from surface smears

Bacteria strain	**Number of CRE isolates**	**ERT**	**EME**	**IMP**
Escherichia coli	3	R	R	R
Acinetobacter baumannii	3	R	R	R
Pantoea sp.	2	R	R	R
Enterobacter cloacae	1	R	R	R
Klebsiella pneumonia	1	R	R	R
Citrobacter freundii	1	R	R	R

The antibiotic susceptibility profiles

A total of 12 CRE isolates from a total of 127 samples from the studied ICUs were interpreted according to the guideline of CLSI (CLSI, 2024). The isolates showed a varying resistance to the ten antibiotics, where the highest resistance was recorded against Ertapenem, Imipenem and Meropenem at 100%, compared to resistance to other antibiotics such as Ceftazidime, Ceftriaxone, Amoxicillin-Clavulanic acid and Cefepime had a resistance rate of (91.7%), while Cefoxitin and Cefotaxime ((75%), then Aztronam had lass resistance compared to other antibiotics that used in this study at rate of (66.7%) (Table 9).

Table 9: Patterns of resistance to individual antibiotics among the CRE isolated from HCWs, and surface smears in ICUs

No. of samples	Name of Bacteria	Antibiotics									
		ETP	IMP	EMP	FOX	AUG	CTR	CFP	CTX	AZT	CAZ
1	*Klebsiella pneumoniae*	R	R	R	R	R	R	R	R	S	R
2	*Acinetobacter baumannii*	R	R	R	R	R	R	R	R	R	R
3	*Acinetobacter baumannii*	R	R	R	R	R	R	R	R	R	R
4	*Acinetobacter baumannii*	R	R	R	R	R	R	R	R	R	R
5	*Escherichia coli*	R	R	R	S	R	R	R	R	S	R
6	*Escherichia coli*	R	R	R	R	R	R	R	S	R	R
7	*Escherichia coli*	R	R	R	R	R	R	R	R	R	R
8	*Pantoea Sp.*	R	R	R	R	R	R	R	R	R	R
9	*Pantoea Sp.*	R	R	R	S	S	S	S	S	S	R
10	*Citrobacter freundii*	R	R	R	R	R	R	R	R	S	S
11	*Enterobacter cloacae*	R	R	R	R	R	R	R	R	R	R
12	*Klebsiella pneumoniae*	R	R	R	R	R	R	R	R	R	R

The HCWs' estimation of nosocomial infection among ICU patients

Since there are no records on the extent of nosocomial infection, we asked HCWs about their estimation of nosocomial infection among inpatients, the results showed that 27.8% of them believe that nosocomial infections is minimal (less than 25%), while 38.9% believed that it could affect up to 50% of patients and 33.3% of HCW reported that it may reach up to 75% of all admitted cases (Table 10).

Table 10: The HCWs' estimation of nosocomial infection among ICU patients in the studied ICUS units

Estimated nosocomial infections in ICUs units	**Number HCWs**	**Percent (%)**
< 25%	10	27.8%
25 – 49%	14	38.9%
50 – 74%	8	22.2%
≥ 75%	4	11.1%
Total	36	100%

HCWs' opinions about the prevention of nosocomial infection by hand hygiene

Asking the HCWs about the role of hand hygiene in preventing nosocomial infections in ICUs units, the results in (Table 11) shows that 13 of them (36.1%) said that hand hygiene prevents higher than 75% of these infections, while 11(30.6%) of them think that hand hygiene prevents between 50 to 74% of these infections in ICU units.

Table 11: HCWs' opinions about the prevention of nosocomial infection by hand hygiene

The percentage of infection prevention by hand hygiene	Number HCWs	Percent (%)
< 25%	5	13.9%
25 – 49%	7	19.4%
50 – 74%	11	30.6%
≥ 75%	13	36.1%
Total	36	100%

DISCUSSION

In intensive care units (ICUs), hospitalized patients are more susceptible to nosocomial pathogens, especially multidrug-resistant bacteria. These bacteria can spread from patient care staff and medical equipment in intensive care units and vice versa, leading to nosocomial outbreaks through cross-infection and/or cross-contamination [(13)]. Therefore, the main objective of this study was to determine the MDR profile of pathogenic bacteria isolated from intensive care units of Toburk Medical Center and their antibiotic susceptibility profiles. Carbapenem-resistant *Enterobacteriaceae* and gram-negative infections have become a critical problem and a significant threat to global health, associated with a high morbidity and mortality rate

[(8, 14-16)]. Results from our study placed the prevalence of carbapenem-resistant among Enterobacteriaceae and gram-negative bacteria isolates in the study location at 66.4%. Our results agree with a study in Egypt, which reported that carbapenem resistance was 62.7% among Enterobacteria [(17, 18)].

The study revealed that the most common microbial isolates from both healthcare workers were Gram-positive organisms such as *Staphylococcus spp,* with a total rate of about 63.9%, while Gram-negative organisms recorded 36.1% as the total percentage. The Gram-negative organisms that were isolated from HCWs were Klebsiella *pneumoniae* (23.1%), *Pantoea Sp.* (23.1%), *Flavimonas oriyzihabitans* (15.4%), *Acinetobacter baumannii* (15.4%), *Escherichia coli* (15.4%) and *Serratia marcescens* (7.7%). This study showed that of the 91 samples collected and identified from surfaces of ICUs, 74.8% yielded microbial isolates. This means that the contamination rate in the studied ICUs was 74.8%, with microbial isolates (both Gram-negative and Gram-positive bacteria). This is a similar study conducted in Ilorin, Nigeria [(19)], which reported a contamination rate of 67.8%, while another study in Maiduguri, Nigeria [(18)], reported a contamination rate of 62.5%. As well as a study in Southeast Nigeria, which reported ICU had a contamination rate of 54.5% [(20)]. The high rate of microbial isolates and the high contamination rate observed in our ICU can be attributed to the semi-open type of operation, with frequent presence and attention from healthcare workers from different units and disciplines, which increases the movement of personnel and activities within the ICU and increases its contamination rate.

The results of this study also showed that of the 36 samples collected and analyzed from healthcare workers in intensive care units, 100% yielded microbial isolates (Gram-negative and Gram-positive bacteria). Previous studies have shown that healthcare workers contribute approximately 20%–40% of infections following cross-contamination within the same unit during clinical procedures [21,22]. This high rate of contamination from inanimate objects and equipment can be inferred from the following: contaminated hands of healthcare workers, ineffective hand washing practices, which frequently serve as a means of cross-contamination, and the lack of proper cleaning procedures for equipment, surfaces, and instruments in the intensive care unit [(20, 23)].

Cross-contamination and transmission of hospital-acquired organisms can occur internally through direct hand contact of healthcare workers with patients and externally through indirect contact with contaminated surfaces, equipment, and instruments in the intensive care units. Regarding antibiotic susceptibility testing, all 12 CRE isolates from healthcare workers and surface smears were highly resistant to Ertapenem, Imipenem, and Meropenem, with a 100% resistance rate. Also, the isolates had a high resistance rate of 91.7% to other antibiotics such as Ceftazidime, Ceftriaxone, Amoxicillin-Clavulanic acid, and Cefepime, while they had a resistance rate of 75% to Cefoxitin and Cefotaxime. The lower resistance was shown to the Aztronam (66.7%) when compared to other antibiotics that were used. In agreement with these findings, previous studies reported 100% resistance of the carbapenem-resistant isolates to multidrug-resistant (defined as non-susceptibility to at least one agent in three different antibiotic groups). These findings are in tandem with existing literature on the subject as reported by Zhang *et al.* (24),Oluwafolajimi & Hilda (25), Fathi *et al.* (26), where all carbapenem-resistant *Enterobacteriaceae* were also resistant to all Cephalosporins tested, and aminoglycosides possessed better activity compared with Fluoroquinolones.

Of the 52 Gram-negative bacteria and Enterobacter infections, 52 (57%) were isolated from HCWs, and surface smears showed apparent resistance to carbapenems. The study showed that *Acinetobacter baumannii* and *Escherichia coli* both had the highest levels of carbapenem resistance isolated from healthcare workers and surface smears (25%). This is consistent with a number of previous studies, which reported that the highest percentage of *Enterobacteriaceae* isolated was reported by Aya et al. (21%) [(27)]. But it was less than that reported by Fathi et al. [(26)], and Deogratius *et al.* [(28)]. In our study, *Klebsiella pneumoniae and Pantoea Sp were* the second most common isolates with (16.7%), followed by *Citrobacter freundii* and *Enterobacter cloacae* (8.3%). While the previous studies reported *Escherichia coli* was the second highest bacterium in other studies, such as [(29)].

The reason for this can be traced back to the mechanism of development of carbapenem-resistance, which is due to the development of extended-spectrum β-lactamases, and Cephalosporins, being β-lactam antibiotics themselves, would therefore be ineffective against CRE infections. Therefore, this study forms an opening to facilitate epidemiological studies. However, in addition to antibiotic combinations for the treatment of CRE, new treatment options using other antibiotics are gradually emerging. However, these all have their own disadvantages, such as drug delivery and resistance. That is why this approach has not been successfully applied in practice. Moreover, the use of new antibiotics leads to different resistance mechanisms and new resistance characteristics may appear. Therefore, the rational use of antibiotics in appropriate combinations is the only promising alternative for infections caused by CRE-like bacteria.

ICU surfaces and HCWs serve as potential reservoirs and vectors for CRE and MDR-GNB transmission. Infection control approaches such as routine environmental surveillance, sink redesign, and antimicrobial surfaces are essential to contain the spread. The findings from this study highlight the widespread presence of carbapenem-resistant *Enterobacteriaceae* (CRE) and other Gram-negative bacteria (GNB) in the ICUs environment. Surfaces such as bed rails and ventilator controls were among the most contaminated. Several isolates from healthcare workers matched environmental strains, supporting the hypothesis that cross-contamination plays a pivotal role in hospital-acquired infection (HAI) dynamics. The high rate of resistance to carbapenems is alarming and reflects the global trend in antimicrobial resistance. Contributing factors include inappropriate antibiotic use, inadequate disinfection, and lapses in hand hygiene. Our study reinforces the urgent need for regular environmental screening programs, strengthened hand hygiene compliance, routine antimicrobial resistance surveillance, and molecular characterization to track resistance genes.

Carbapenem is one of the antibiotics that offer broad-spectrum activity and is used as a last-line therapy for multidrug-resistant bacteria. The treatment of infections caused by drug-resistant bacteria is sometimes impossible and may lead to unexpected or bad complications. Antimicrobial resistance increases the cost of health care and the possibility of complications. Without effective antimicrobials for the prevention and treatment of CRE infections, medical procedures become very high-risk. The major worrisome aspect is that treatment of the infections caused by these multidrug organisms is extremely difficult, which may result in high mortality rates and healthcare costs.

The analytical results presented in this study provide insight into the epidemiology of carbapenem-resistant *Enterobacteriaceae* and other Gram-negative bacteria in the healthcare setting studied. In addition, we demonstrated how routinely collected microbiology laboratory data can be used to gain insight into infectious disease trends and emerging threats; however, this study is not without limitations. First, we did not report the molecular epidemiology of carbapenem resistance mechanisms in the study population. This is important to understand the molecular basis for the emergence of these observed resistance characteristics. Second, we also did not report hospital-level carbapenem resistance rates. The reasons for this were that the required data could not be collected due to difficulties in collecting the necessary data, a lack of sufficient collaboration, and the absence of a centralized electronic health information management system at the study site. These data, if available, would provide insight into clinical risk factors associated with carbapenem resistance in the study population, which could inform health promotion efforts. Finally, due to the small sample size, we were unable to perform organism-level analyses to determine trends in carbapenem resistance among bacterial species and investigate possible differences or associations.

CONCLUSIONS

This study provides critical evidence of environmental and occupational contamination with CRE and other resistant Gram-negative bacteria in ICU settings. The results underline the necessity of comprehensive infection control strategies, regular decontamination protocols, and continuous education of healthcare workers to mitigate the spread of multidrug-resistant organisms.

The researchers recommended that to improve diagnosis by providing specialized laboratories for the detection of CRE, to increase medical awareness about CRE by well training healthcare professionals on the cautious use of antibiotics. Also, to enhance prevention policies by implementation of strict infection control policies, such as sterilizing medical instruments and isolating infected patients, and to conduct scientific research with a focus on the development of new antibiotics and monitoring the genetic patterns of resistant bacteria**.**

REFERENCES

1. World Health Organization. WHO bacterial priority pathogens list, 2024: bacterial pathogens of public health importance to guide research, development and strategies to prevent and control antimicrobial resistance. Geneva: World Health Organization; 2024.
2. Chaudhary R, Bhatta S, Pant C, Phuyal N, Shrestha L. Multidrug resistance gram-negative bacteria in intensive care unit of tertiary care hospital: a descriptive cross-sectional study. JNMA J Nepal Med Assoc. 2024;62(277):582-6. PubMed PMID: 40655052.
3. Aldali HJ, Khan A, Alshehri AA, Aldali JA, Meo SA, Hindi A, et al. Hospital-acquired infections caused by carbapenem-resistant Enterobacteriaceae: an observational study. Microorganisms. 2023;11(6):1595.
4. World Health Organization. Global antimicrobial resistance and use surveillance system (GLASS) report: 2022. Geneva: World Health Organization; 2022.
5. Kuloglu TO, Unuvar GK, Cevahir F, Kilic AU, Alp E. Risk factors and mortality rates of carbapenem-resistant Gram-negative bacterial infections in intensive care units. J Intensive Med. 2024;4(3):347-54. PubMed PMID: 39035617.
6. Ibrahim K, Thwood D, ELgheriani H, Salem M, Elgadiym Z, Zaghdani A, et al. Prevalence of multi-drug resistant bacteria in intensive care units at Tripoli University Hospital, Tripoli, Libya. Libyan J Med. 2024;19(1):2348235.
7. Elramli A, Almoghraby R, Mohamed F, Zaghdani A, Asolimany S, Arfaoui A, et al. Molecular characterization of multidrug-resistant Klebsiella pneumoniae isolated from some hospitals in Benghazi, Libya. AlQalam J Med Appl Sci. 2024:590-6.
8. Barco L, Belluco S, Roccato A, Ricci A. Escherichia coli and Enterobacteriaceae counts on poultry carcasses along the slaughter processing line, factors influencing the counts and relationship between visual faecal contamination of carcasses and counts: a review. EFSA Support Publ. 2014;11(8):636E.
9. Otter JA, Yezli S, Salkeld JA, French GL. Evidence that contaminated surfaces contribute to the

transmission of hospital pathogens and an overview of strategies to address contaminated surfaces in hospital settings. Am J Infect Control. 2013;41(5 Suppl):S6-11. PubMed PMID: 23622751.
10. Alquraishi FE, Al-Saadi ZN, Al-Azzawi JAT. Determination of multi-drug resistant Klebsiella pneumoniae isolated from UTI patients in Wasit province, Iraq. Int J Health Sci. 2022;6(S6):7937-49.
11. Kachrimanidou M, Tsintarakis E. Insights into the role of human gut microbiota in Clostridioides difficile infection. Microorganisms. 2020;8(2).
12. Galvin S, Dolan A, Cahill O, Daniels S, Humphreys H. Microbial monitoring of the hospital environment: why and how? J Hosp Infect. 2012;82(3):143-51. PubMed PMID: 23022372.
13. Nordmann P, Poirel L, Walsh TR, Livermore DM. The emerging NDM carbapenemases. Trends Microbiol. 2011;19(12):588-95. PubMed PMID: 22078325.
14. Morrill HJ, Pogue JM, Kaye KS, LaPlante KL. Treatment options for carbapenem-resistant Enterobacteriaceae infections. Open Forum Infect Dis. 2015;2(2):ofv050. PubMed PMID: 26125030.
15. Amer WH, Khalil HS, Abd El Wahab MA. Risk factors, phenotypic and genotypic characterization of carbapenem resistant Enterobacteriaceae in Tanta University Hospitals, Egypt. Int J Infect Control. 2016;12(2).
16. Tamma PD, Goodman KE, Harris AD, Tekle T, Roberts A, Taiwo A, et al. Comparing the outcomes of patients with carbapenemase-producing and non-carbapenemase-producing carbapenem-resistant Enterobacteriaceae bacteremia. Clin Infect Dis. 2017;64(3):257-64. PubMed PMID: 28013264.
17. Damaceno Q, Iquiapaza R, Oliveira A. Comparing resistant microorganisms isolated from patients and environment in an intensive care unit. Adv Infect Dis. 2014;4:30-5.
18. Anokwute II, Onwudiwe RU, Kalu E, Madubuko CG, Egbulem CT, Eluchie EC. Determination of the common microorganisms present in the intensive care unit of Federal Teaching Hospital Owerri, Southeast Nigeria: a prospective, descriptive cross-sectional study. Niger Postgrad Med J. 2025;32(1):19-24. PubMed PMID: 40091467.
19. Abubakar S, Mohammed M, Habiba J, Yusuf S, Goni B, Josiah D. Spectrum of bacterial isolates among intensive care units patients in a tertiary hospital in northeastern Nigeria. Indian J Sci Resour Technol. 2014;2(6):42-7.
20. Huang SS, Datta R, Platt R. Risk of acquiring antibiotic-resistant bacteria from prior room occupants. Arch Intern Med. 2006;166(18):1945-51. PubMed PMID: 17030826.
21. Hayden MK, Blom DW, Lyle EA, Moore CG, Weinstein RA. Risk of hand or glove contamination after contact with patients colonized with vancomycin-resistant enterococcus or the colonized patients' environment. Infect Control Hosp Epidemiol. 2008;29(2):149-54. PubMed PMID: 18179370.
22. Cordeiro ALAO, Oliveira MMC, Fernandes JD, Barros CSMA, Castro LMC. Equipment contamination in an intensive care unit. Acta Paul Enferm. 2015;28(2):160-5.
23. Sax H, Allegranzi B, Uçkay I, Larson E, Boyce J, Pittet D. 'My five moments for hand hygiene': a user-centred design approach to understand, train, monitor and report hand hygiene. J Hosp Infect. 2007;67(1):9-21. PubMed PMID: 17719685.
24. Zhang Y, Guo LY, Song WQ, Wang Y, Dong F, Liu G. Risk factors for carbapenem-resistant K. pneumoniae bloodstream infection and predictors of mortality in Chinese paediatric patients. BMC Infect Dis. 2018;18(1):248. PubMed PMID: 29855274.
25. Adesanya OA, Igwe HA. Carbapenem-resistant Enterobacteriaceae (CRE) and gram-negative bacterial infections in south-west Nigeria: a retrospective epidemiological surveillance study. AIMS Public Health. 2020;7(4):804-15. PubMed PMID: 33294483.
26. Fathi S, Hassan H, Alfageih L. Prevalence of carbapenem-resistant Enterobacteriaceae isolated from clinical samples in Medical Tobruk Center. Razi Med J. 2025:126-36.
27. Al-Baz AA, Maarouf A, Marei A, Abdallah AL. Prevalence and antibiotic resistance profiles of carbapenem-resistant Klebsiella pneumoniae isolated from Tertiary Care Hospital, Egypt. Egypt J Hosp Med. 2022;88(1):2883-90.
28. Okoche D, Asiimwe BB, Katabazi FA, Kato L, Najjuka CF. Prevalence and characterization of carbapenem-resistant Enterobacteriaceae isolated from Mulago National Referral Hospital, Uganda. PLoS One. 2015;10(8):e0135745. PubMed PMID: 26270465.
29. Bhandari A, Khatiwada S, Sharma A, Aryal SC, Shrestha R, Bimali NK, et al. Prevalence of drug resistant Enterobacteriaceae in a Nepalese tertiary care hospital. PLOS Glob Public Health. 2024;4(1):e0000858.