ISBN: 978-628-96613-0-9. ISSN: 2414-6390. Digital Object Identifier: https://dx.doi.org/10.18687/LEIRD2024.1.1.883

Production and coproduction of carbapenemases in Enterobacterales isolated from patients in a Peruvian hospital treated during the COVID-19 pandemic.

David C. García [1], Luis A. Cabanillas-Chirinos[2], De La Cruz-Noriega, M.[3], Karen Diaz del Aguila [2], Santiago M. Benites

[1] Facultad de Ciencias de la Salud, Universidad César Vallejo, Trujillo 13001, Peru
dcgarciac@ucvvirtual.edu.pe
[2] *Programa de Investigación Formativa, Universidad César Vallejo, Trujillo 13001.*
lcabanillas@ucv.edu.pe, kdiazd@ucv.edu.pe*,*
[3] Institutos y Centros de Investigación.Universidad Cesar Vallejo, Trujillo 13001, Peru; mdelacruzn@ucv.edu.pe,
[4] Vicerrectorado de Investigación, Universidad Autónoma del Perú, Lima 15842, Peru santiago.benites@autonoma.pe

Abstract– In Peru, from May 2021 to March 2022, hospital care was restricted due to the second and third wave of the COVID-19 pandemic. During this period a large number of cases of acute respiratory infections increased. With increasing resistance to antibacterials, the search for and identification of carbapenemase co-producing Enterobacteriaceae is of paramount therapeutic importance because it limits the use of available antibiotics. Objective: To determine the Enterobacterales species that present carbapenemase: KPC, NMD and OXA 48 in patients treated in a COVID hospital in Trujillo-Peru, during the COVID-19 pandemic. Methodology: In this descriptive, observational study, biological samples were collected from patients treated in general medicine, emergency, and intensive care units - ICU of Hospital IV "Víctor Lazarte Echegaray" for bacterial identification and sensitivity to carbapenems. removed AutoScan-4 automated system, and the rapid diagnostic immunochromatographic tests RESISIT-3 OKN K-SET were used to determine the type of carbapenemase present in each culture. Results: Of a total of 134 Enterobacterales cultures, **Klebsiella pneumoniae** ***is the one with the highest resistance to imipenem and meropenem; the highest prevalence of resistance for both carbapenems was obtained in the ICU (n=22; 54.6%) compared to that obtained in general and emergency medicine. Conclusions: Most*** **K. pneumoniae** ***cultures showed KPC-type carbapenemase, and no bacteria were found capable of co-producing two or more carbapenemases.***

Keywords-- Enterobacterales; Carbapenem resistance; co-production of carbapenemases.

I. Introduction

At a clinical level, Enterobacterales acquire great importance because they cause a wide variety of diseases in humans, through hospital-acquired infections and community infections such as infections in the urinary tract, respiratory tract, bloodstream infections, and infections of exposed wounds; It is currently becoming evident that some Enterobacteriaceae species are capable of acquiring resistance to broad-spectrum antibiotics [1-9].

The World Health Organization (WHO), in 2022, presented a global report, that mentions that, in developed countries, 7 out of every 100 hospitalized patients will contract at least one nosocomial infection during their hospitalization, while in underdeveloped and developing countries, the figure can rise to 15 out of every 100 patients [10]. In recent years it has been observed that a progressive increase in infections caused by resistant and multi-resistant bacteria is causing ineffectiveness in antimicrobial treatments [11]. The Spanish Society of Infectious Diseases and Clinical Microbiology SEIMC (2022) mentions that by not considering antimicrobial resistance, it is possible that by the year 2050, more than 10 million deaths will occur [12].

The Pan American Health Organization/World Health Organization - PAHO/WHO, in October 2021, issued an epidemiological alert throughout Latin America and the Caribbean, which mentioned the increase in resistance to antibacterials due to new mixtures of carbapenemases in Enterobacterales during the COVID-19 pandemic and relates this increase in resistance with the uncontrolled use of broad-spectrum antibiotics in patients with COVID-19 (PAHO/WHO, 2021) [13]. For Yañez (2021) the COVID-19 caused a dramatic increase in the number of patients hospitalized in the ICU area, evidencing the irrational use of antimicrobials without scientific pharmacological support, which increased in the presence of resistant microorganisms [3, 14].

Beta-lactams are considered time-dependent antibiotics because they have slow bactericidal activity since their prolonged exposure helps to eliminate bacteria even more [8,15,16]. On the contrary, beta-lactamases are bacterial enzymes that inhibit the mechanism of action of beta-lactam antibiotics through the hydrolysis process of the antibiotic [2]; and within beta-lactams, we have carbapenems, which have a broad spectrum of antibacterial activity and are used for the control and elimination of gram-positive and gram-negative microorganisms [17]. For this reason, they are considered the most innovative and reliable medicine to treat infections, because they are safer than other medicines and have few side effects on people [8,15].

Imipenem, meropenem, ertapenem, and doripenem, until 2010, have been the carbapenems most used in the clinic during the treatment of infectious diseases caused by bacteria [18]; and of these, imipenem and meropenem stand out as they are the antibiotics most used in the treatment. of many respiratory, intra-abdominal, and nosocomial infections, effectively combating infections caused by *Pseudomonas aureginosa* [9,19]. Initially, the appearance of carbapenemases in patients with bacterial infections was rare. Currently, the increase in carbapenemases is part of the defense mechanism used by bacteria; and especially in Enterobacterales, is generating concern in the therapeutic field since it has been observed that resistance to carbapenems implies resistance to other β-lactams [7,17]. Like other ß-lactams, Imipenem and Meropenem have the function of inhibiting the synthesis of the bacterial wall, but they differ in the mechanism of action since Imipenem has an affinity for Opr D2 porins, while Meropenem has a greater affinity for PBPs (penicillin-binding proteins) which are β-lactam binding proteins [5,18,19].

Multidrug-resistant GNB constitutes a health problem of global relevance, for this reason, PAHO/WHO indicates the need to search for new antibiotics to combat bacteria resistant to carbapenems or producers of extended-spectrum ß-lactamases.5 Initially, the Carbapenem resistance was focused on *P. aeruginosa* and *Acinetobacter baumannii*, but in recent decades *Klebsiella pneumoniae* was also included [20]. In Peru, the Ministry of Health (2022) identified two strains of *K. pneumoniae*, with co-production of KPC+NDM type carbapenemases, a strain of *A. baumannii* coproducing NDM+OXA-58 and an unusual strain of *Escherichia coli* coproducing NDM+OXA-48 [21].

Various studies show that, within hospitals, the Intensive Care Unit (ICU) is an area where the risk is high for antimicrobial resistance to occur due to the high rate of antibiotic use [22,23]. The percentage of bacteria, that present resistance to antibiotics, isolated from the ICU is almost double compared to other clinical areas [24]. The presence of GNB resistant to antibiotics has led to new and different containment routes being proposed to combat them, for this reason, it is important to carry out work that allows us to know and identify the appearance and frequency of bacterial species resistant to carbapenems with ability to coproduce more than one beta-lactamase. The present study aims to determine the prevalence of Enterobacterales producing KPC, NDM, and OXA-48 in patients treated at a level IV hospital during the COVID-19 pandemic.

II. Material and methods

A. Design of the study

A descriptive and observational study was carried out based on the isolation, identification, and sensitivity to carbapenems and carbapenemase production tests carried out on cultures obtained from patients treated in the general medicine, emergency, and ICU area of Hospital IV "Víctor Lazarte Echegaray", located in Trujillo, Peru; during May 2021 to March 2022, a period in which the second and third waves of the COVID-19 pandemic occurred in Peru, taking into account that this hospital was considered a reference hospital for COVID treatment.

B. Population and sample

The population sample consisted of 134 Enterobacteriaceae cultures isolated from patients treated in the general medicine, emergency, and ICU area of Hospital IV "Víctor Lazarte Echegaray" between May 2021 and March 2022; and the sociodemographic characteristics of the patients are presented in the results item.

C. Collection and Processing of biological samples

Biological samples of bronchial secretion, aseptic urine, blood, wound secretion, and other biological fluids; were collected and processed following the guidelines of the Laboratory Procedures Manual: Local Laboratories I: Local Laboratories II of the Ministry of Health/National Institute of Health [25].

D. Selection, identification, and Sensitivity of Bacteria

Colonies that developed on Mac Conkey agar plates were selected for identification and susceptibility testing using the automated MicroScan™ AutoScan-4 system and Neg Entero Combo Panel Type 72, from Siemens Healthcare Diagnostics - USA. which contains carbapenems: imipenem, meropenem, and ertapenem which can be used in the treatment of multidrug-resistant GNB present in urine and systemic samples. The bacterial identification and sensitivity procedure included making a bacterial suspension and transferring 100 μL to the wells of the panels, which were incubated for 16 to 18 hours at 35°C. The Enterobacterales cultures that presented Minimum Inhibitory Concentration MICs values > 1 mg/l for imipenem and meropenem were considered resistant to these antibiotics, and were separated to perform the carbapenemase production test; Furthermore, all the mentioned procedures were performed according to the manufacturer instructions [25-29].

E. Identification of Carbapenemases

The identification of the carbapenemases KPC-type, NDM, and OXA-48, was carried out using a rapid diagnostic immunochromatographic technique RESISIT-3 OKN K-SET manufactured by the company Coris BioConcept, which has a sensitivity of 96% and 100% specificity for the detection of type carbapenemases KPC, NDM, and OXA-48 [30,31].

F. Data Collection and Analysis

The data obtained were recorded in the work panels' worksheets and then transferred to the LabPro software version 4.43 for the determination of the "biotype"; results with high probabilities (≥85%) were considered reliable, and those with low probability (<85%) were considered "not confirmed" [28]. In this study, no additional testing was performed to confirm low-probability identifications and the antibiotic resistance profile was interpreted according to the breakpoints recommended by the Clinical and Laboratory Standards Institute [26,27]. To understand the results, graphs and tables were generated in the Microsoft Excel program.

III. Results

In Table I, the 134 cultures consisted of 62 cultures from general medicine patients, 31 cultures from emergency patients, and 41 cultures from the ICU. Biological samples included bronchial secretion, aseptic urine, blood, central venous catheter, wound secretion, and other biological fluids. As exclusion criteria, cultures that do not belong to the order Enterobacterales and non-viable cultures were taken into consideration, and as inclusion criteria, viable axenic cultures from samples from patients treated in the general medicine, emergency; and ICU areas were considered.

TABLE I
SOCIODEMOGRAPHIC CHARACTERISTICS OF GENERAL MEDICINE, EMERGENCY, AND ICU PATIENTS TREATED AT HOSPITAL IV "VÍCTOR LAZARTE ECHEGARAY", FROM MAY 2021 TO MARCH 2022.

General information	n = 134 (100,0 %)		
	General medicine n = 62	Emergency n = 31	ICU n = 41
Age average (in years)	38 (20 – 58)	48 (25 -56)	52 (37 - 79)
Female	30	18	17
Male	32	13	24
Bacterial Colonization	6	0	0
Bacterial Infection	56	31	31
COVID-19 Positives	0	6	27
COVID-19 Negatives	62	25	14

In Table II, shows the frequency distribution of isolated cultures that present resistance to carbapenems, according to the type of sample obtained in the area of general medicine, emergency and ICU. In the general medicine and emergency area, the vast majority of cultures were isolated from aseptic urine samples (n = 34; 54.8 and n = 20; 55.6%) while in the ICU the majority of cultures were isolated from samples bronchial secretion (n=27; 65.9%).

TABLE II
FREQUENCY OF BACTERIAL CULTURES RESISTANT TO IMIPENEM AND MEROPENEM, BY TYPE OF SAMPLE OF PATIENTS TREATED IN THE HOSPITAL "VÍCTOR LAZARTE ECHEGARAY", FROM MAY 2021 AND MARCH 2022.

Type of Sample	General medicine Isolated culture n (%)	Emergency Isolated culture n (%)	UCI Isolated culture n (%)
Aseptic urine	34 (54.8)	20 (64.5)	14 (34.1)
Bronchial secretion	14 (22.6)	6 (19.4)	27 (65.9)
Blood culture	3 (4.8)	4 (12.9)	0 (0.0)
Surgical wound	6 (8.6)	0 (0.0)	0 (0.0)
Otros	5 (8.1)	1 (3.2)	0 (0.0)
Total	70 (100)	31 (100)	41 (100)

In Table III, shows the distribution and frequency of isolated bacterial cultures resistant to imipenem and meropenem, where it observed that *Klebsiella pneumoniae* and *Escherichia coli* cultures predominated in the samples obtained from general medicine, emergency, and medical patients. ICU. The enterobacteria that present the greatest resistance to imipenem and meropenem is *K. pneumoniae*, and the frequency of its resistance to the two carbapenems in the ICU (n=22; 54.6%) is higher compared to that obtained in general medicine and emergency; while *E. coli* presents greater resistance only to imipenem in general medicine (n=11; 19.3%) and while resistance to imipenem and meropenem in the emergency area (n=8; 50.0%) and ICU (n=5;12.5%) is greater than these two carbapenems. On the other hand, it is also shown, with less or no frequency, in a minority of bacterial cultures resistant to imipenem and/or meropenem in each hospital environment.

TABLE III
DISTRIBUTION OF BACTERIA RESISTANT TO IMIPENEM AND/OR MEROPENEM IN PATIENTS TREATED IN THE HOSPITAL "VÍCTOR LAZARTE ECHEGARAY", FROM MAY 2021 AND MARCH 2022.

Area/Isolated Bacteria	Isolated culture n (%)	Resistant to Imipenem n (%)	Resistant to Meropenem n (%)	Resistant to Imipenem y Meropenem n (%)
General medicine				
Klebsiella pneumoniae	32 (51.6)	29 (50.9)	2 (40.0)	1 (50.0)
Escherichia coli	12 (19.4)	11 (19.3)	0 (0.0)	0 (0.0)
Enterobacter cloacae	5 (8.1)	5 (8.8)	2 (40.0)	1 (50.0)
Proteus mirabilis	4 (6.5)	3 (5.3)	1 (20.0)	0 (0.0)
Morganella morganii	1 (1.6)	1 (1.8)	0 (0.0)	0 (0.0)
Serratia odorifera	1 (1.6)	1 (1.8)	0 (0.0)	0 (0.0)
Klebsiella oxytoca	3 (4.8)	3 (5.3)	0 (0.0)	0 (0.0)
Klebsiella ozaenae	1 (1.6)	1 (1.8)	0 (0.0)	0 (0.0)
Burkholderia cepacia complex	1 (1.6)	1 (1.8)	0 (0.0)	0 (0.0)
Kluyvera ascorbata	1 (1.6)	1 (1.8)	0 (0.0)	0 (0.0)
Proteus penneri	1 (1.6)	1 (1.8)	0 (0.0)	0 (0.0)
Total	62 (100.0)	57 (100.0)	5 (100.0)	2 (100.0)
Emergency				
Klebsiella pneumoniae	9 (29.0)	9 (30.0)	5 (25.0)	5 (31.3)
Escherichia coli	9 (29.0)	9 (30.0)	8 (40.0)	8 (50.0)
Enterobacter cloacae	3 (9.7)	3 (10.0)	0 (0.0)	0 (0.0)
Enterobacter aerogenes	5 (16.1)	4 (13.3)	4 (20.0)	0 (0.0)
Proteus mirabilis	3 (9.7)	3 (10.0)	2 (10.0)	2 (12.5)
Morganella morganii	1 (3.2)	1 (3.3)	1 (5.0)	1 (6.3)
Serratia odorifera	1 (3.2)	1 (3.3)	0 (0.0)	0 (0.0)
Total	31 (100.0)	30 (100.0)	20 (100.0)	16 (100.0)
ICU				
Klebsiella pneumoniae	22 (53.7)	22 (53.7)	22 (56.4)	22 (56.4)
Escherichia coli	6 (14.6)	6 (14.6)	5 (12.8)	5 (12.8)
Enterobacter cloacae	1 (2.4)	1 (2.4)	1 (2.6)	1 (2.6)
Enterobacter aerogenes	1 (2.4)	1 (2.4)	1 (2.6)	1 (2.6)
Proteus mirabilis	1 (2.4)	1 (2.4)	1 (2.6)	1 (2.6)
Morganella morganii	1 (2.4)	1 (2.4)	1 (2.6)	1 (2.6)

Klebsiella oxytoca	3 (7.3)	3 (7.3)	3 (7.7)	3 (7.7)
Burkholderia cepacia complex	3 (7.3)	3 (7.3)	3 (7.7)	3 (7.7)
Serratia marcescens	2 (4.9)	2 (4.9)	2 (5.1)	2 (5.1)
Citrobacter freeundii	1 (2.4)	1 (2.4)	0 (0.0)	0 (0.0)
Total	41 (100.0)	41 (100.0)	39 (100.0)	39 (100.0)

In Table IV, shows the different bacteria groups and the carbapenemase KPC, NDM, and OXA-48, which they produce; These bacteria can cause nosocomial infections. The majority of *K. pneumoniae* cultures have KPC-type carbapenemases (general medicine, n = 23; emergency, n = 6 and ICU, n = 22). The other bacterial groups have at least the presence of one type of carbapenemase in smaller quantities: No bacteria were found that have 2 or more types of carbapenemase in common.

TABLE IV
DISTRIBUTION OF BACTERIA, BY TYPE OF CARBAPENEMASE IT PRODUCES, ISOLATED IN PATIENTS TREATED IN THE HOSPITAL "VÍCTOR LAZARTE ECHEGARAY", FROM MAY 2021 TO MARCH 2022.

Area/Isolated Bacteria	cultures with carbapenemasa n (%)	Tipo de carbapenemasa		
		KPC	NDM	OXA - 48
General medicine				
Klebsiella pneumoniae	24 (72.7)	23	1	0
Escherichia coli	5 (15.2)	0	3	2
Klebsiella oxytoca	2 (6.1)	2	0	0
Klebsiella ozaenae	1 (3.0)	1	0	0
Kluyvera ascorbata	1 (3.0)	0	1	0
Total	33 (100.0)	26	5	2
Emergency				
Klebsiella pneumoniae	8 (33.3)	6	2	0
Escherichia coli	7 (29.4)	0	2	5
Enterobacter cloacae	1 (4.2)	0	0	1
Enterobacter aerogenes	4 (16.7)	0	2	2
Proteus mirabilis	3 (12.5)	0	2	1
Morganella morganii	1 (4.2)	0	1	0
Total	24 (100.0)	6	9	9
ICU				
Klebsiella pneumoniae	22 (56.4)	22	0	0
Escherichia coli	6 (15.4)	0	4	2
Enterobacter cloacae	1 (2.6)	0	1	0
Enterobacter aerogenes	1 (2.6)	0	0	1
Proteus mirabilis	1 (2.6)	0	0	1
Morganella morganii	1 (2.6)	0	1	0
Klebsiella oxytoca	3 (7.7)	3	0	0
Burkholderia cepacia complex	3 (7.7)	0	1	2
Serratia marcescens	1 (2.6)	0	0	1
Total	39 (100.0)	25	7	7

IV. DISCUSSION

The latest advances in robotics and computing have contributed to the development of automated equipment that helps in the diagnosis of diseases; Microbiology laboratories are not exempt from the use of new technologies for the study of susceptibility to antimicrobials intending to simplify the process [32,33]. The hospital "Víctor Lazarte Echegaray", located in Trujillo-Peru, has the automated MicroScan™ AutoScan-4 system and the LabPro software for bacterial identification and sensitivity; This automated system allows the results to be categorized based on the CMI cut-off points recommended by the CLSI; where bacteria with MICs > 1mg/l for Imipenem and Meropenem are considered resistant to these antibiotics and are suspected of producing carbapenemases [20,26,27,29].

Between May 2021 and March 2022, during the second and third waves of the COVID-19 pandemic, 134 biological samples from general medicine, emergency, and ICU at the Hospital-COVID "Víctor Lazarte Echegaray" were analyzed; it was found that the sample of aseptic urine and bronchial secretion are more frequent (see Table II). The high number of bronchial secretion samples present in the ICU (n=27; 65.9%) is associated with the increase in patients treated with acute respiratory infections during the COVID-19 health emergency since during 2021 and 2022, priority was given care to patients suspected of having the SARS-CoV-2 virus; This information obtained agrees with what was reported by Tranche et al. (2021) explained that during the first to third waves of the COVID-19 pandemic, it was people with acute respiratory infections who sought care in hospitals and whose care was prioritized, and secondly, patients who had a serious or chronic illness [35]. This criterion was adopted worldwide, during the time of the pandemic, and Hospital IV "Víctor Lazarte Echegaray" adopted the provisions of the Ministry of Health who established a restriction on care with priority care for patients diagnosed with pneumonia associated with COVID-19, in addition during the diagnosis and treatment of the COVID-19 disease, a culture of bronchial secretion was performed, to diagnose or rule out the appearance of a bacterial infection associated with COVID-19.

In Table III, shows the distribution and frequency of the bacterial cultures isolated and resistant to imipenem and meropenem, observing that the cultures of *K. pneumoniae* and *E. coli* are the cultures that were isolated most frequently in the samples obtained from the patients of general medicine, emergency, and ICU. The enterobacteria with the highest frequency is *K. pneumoniae*, the frequency obtained in the ICU (n=22; 53.7%) was higher than that observed in the area of general medicine and emergency; These data obtained exceed those reported by Spiess et al. (2022), who found that the prevalence of *K. pneumoniae* 39.2%, followed by *E. coli* with 34.2%;9 *K. pneumoniae*, resistant to imipenem, is the most frequently isolated Enterobacteriaceae in general medicine samples (n=29; 50.9%); The frequency of this bacteria is also high in the ICU sample where it presents resistance to imipenem and meropenem (n=22; 53.7% and n=22; 56.4%); In the case of *E. coli*, it was found that this bacteria present resistance to imipenen and meropenem, in the emergency area (n=9; 30.0% and n=8; 40.0%). At the level of South America, these results are much higher than those found by Bravo, Juan

(2020) who reported resistance of 21.2% for imipenem and 16.80% for meropenem;36 and what was reported by Duque Nossa and Varona Alzate (2022) who found that more than a third of patients hospitalized in the ICU presented resistance to AmpC-type beta-lactamase and extended-spectrum beta-lactamase (ESBL);37 While Quintero and Varón, (2022), who indicate that 36.8% of *K. pneumoniae* isolates are multidrug-resistant [38]; thus reaffirming that this type of infection is a major public health problem due to its association with mortality rates greater than 50%; and that in the ICU area, there is a high risk of developing this type of infection because it originates from the same microbiota of the patients and their hospital stay. The greatest resistance to the two carbapenems (imipenem and meropenem) occurs in the samples obtained from ICU patients (n=22; 56.4%), at the national level, these results show an increase compared to what was found by Vicente Castro (2016) in their study carried out in Arequipa, reported that 50% of the *K. pneumoniae* cultures, from UCI - Arequipa Clinic, present resistance to β-lactams (except Carbapanems) [39]; and from what was reported in Ica by Hernandez Bonifaz (2021) who reported that 8.7% of GNB presented resistance to Gentamicin and 7.9% to Amoxicillin + Clavulanic Acid [40]. Although this value obtained is indeed very high, it should be mentioned that all precautions and measures are taken to avoid obtaining crops with false resistance since it is known that on some occasions automated systems can offer anomalous and/or erroneous results, directly related to technical problems; Fernandez et al. (2000) mention that the factor that has been most related to false resistance to imipenem is its low instability since the concentration of imipenem in commercial sensitivity panels can decrease up to 50% due to time factors and storage conditions [32]. For this reason, in the present work, inputs or sensitivity panels were used for a period of no more than six months as indicated by the manufacturer's specifications.

Enterobacterales have various hydrolyzing enzymes as part of their defense mechanism, including KPC (*K. pneumoniae* carbapenemase), NDM (New Delhi Metallo-beta-lactamase), OXA (Oxacillinases), IMP (Imipenemases); and VIM (Verona Integron Encoded Metallo-β-lactamase) which occurs most frequently worldwide [21,34]. With the COVID-19, in South American countries, there was an increase in the incidence of carbapenem resistance, which can be attributed to the increase in the indiscriminate use of broad-spectrum antibiotics [19,41]. The expression of two or more carbapenemases in Enterobacteriaceae is becoming common in Latin America and poses a great diagnostic and therapeutic challenge because it limits the use of all β -lactams currently available. In Ecuador at the beginning of 2021, alerts were raised about the first isolations of *K. pneumoniae* co-producing KPC and NDM and cultures of *E. coli* co-producing KPC and OXA-48, in a third-level national reference hospital. In Peru, at the end of 2021, the appearance of two strains of *K. pneumoniae* was reported, with double production of carbapenemases of the unusual type (KPC + NDM) and an unusual strain of *E. coli* with double production of carbapenemases (NDM+OXA-48) [13,14,21]. In Table IV, shows the type of carbapenemase KPC, NDM, and OXA-48 present in bacterial cultures resistant to carbapenems (imipenem and meropenem) where the results obtained show that in the cultures studied the three types of carbapenemases KPC, NDM, and OXA-48 are present, but in no crop does it co-produce more than one carbapenemase; Furthermore, this Table IV, shows that the enterobacteria *K. pneumoniae* are the one that presents the greatest resistance to carbapenemases in the three environments from which it was isolated and at the level of South America, Caicedo et al. (2023), carried out a study in Ecuador, where the frequency of resistance to carbapenems in hospitalized patients was 60% [42]; these results are very similar to those obtained in the present study where the frequency in general medicine was 72.7%, in an emergency it was 33.3% and in ICU it was 56.4%. Likewise, *K. pneumoniae* presents a greater number of strains with KPC-type carbapenemase in bacterial isolates from general medicine (n = 23), emergency (n = 6), and ICU (n = 22); These results differ and are much superior to those obtained by Guerra et al. (2020) reported that 24% of *K. pneumoniae* cultures present the KPC-type carbapenemase [43].

The results regarding the frequency of the type of carbapenemase found in the different bacterial groups (carbapenemase class A) differ from a similar study carried out in 2019 by the National Institute of Health (INS) of Peru, which reported that 185 strains have class A, B and D carbapenemase enzymes; class B being the most frequent; They also mention genes such as KPC, NDM, IMP, VIM, OXA-24 [24,25]; and the information obtained in the present study is of utmost importance since the Infectious Diseases Society of America recommends avoiding the empirical use of antibiotics when bacterial resistance exceeds 20%, and also shows that the problem of drug resistance in health care units intensive care ICU, is latent and constantly increasing, so stricter measures must be taken to face this problem, which is increasingly larger worldwide, putting the health of patients at risk [20,34,35].

V. CONCLUSION

In this study, between 5 to 9 different species of Enterobacteriaceae resistant to the carbapenems imipenem and meropenem were identified in samples obtained from general medicine, emergency, and ICU patients at the "Víctor Lazarte Echegaray" COVID-Hospital during the COVID-19 pandemic.

Klebsiella pneumoniae and *Escherichia coli* are the enterobacteria that present the highest frequency of resistance to imipenem and meropenem, where the greatest number of resistant cultures was isolated in the samples obtained from ICU patients.

Most *Klebsiella. pneumoniae* cultures showed KPC-type carbapenemase.

No bacteria were found capable of co-producing two or more carbapenemases.

ACKNOWLEDGMENTS

The authors would like to thank the support received by the Hospital IV "Víctor Lazarte Echegaray" during the execution of this research.

REFERENCES

[1] Armijos JAG, Cano ICM, Coronel AAR, Clavijo ACS. Prevención de infecciones asociadas a la atención en salud: revisión sistemática. jah 2021. https://doi.org/10.37958/jah.v0i0.83.
[2] León-Luna D, Fajardo-Loyola A, Yareta-Yareta J, Burgos-Espejo A, Peralta-Siesquen C, Galarza-Pérez M, et al. Caracterización molecular de enterobacterias multirresistentes en dos departamentos de la selva peruana. Biomedica 2021;41:180-7. https://doi.org/10.7705/biomedica.5720.
[3] Vinicoff G, Díaz N, Zubeldía Brenner L, Ezcurra MC, Farina J. Epidemiología de las infecciones asociadas al cuidado de la salud y consumo de antibióticos en pandemia covid 19. Pub cient HCANK 2022;1.
[4] Struthers K. Microbiología clínica. Editorial El Manual Moderno; 2018.
[5] Lepe JA, Martínez-Martínez L. Mecanismos de resistencia en bacterias gramnegativas. Med Intensiva 2022;46:392-402. https://doi.org/10.1016/j.medin.2022.02.004.
[6] Sacsaquispe-Contreras R, Bailón-Calderón H. Identification of carbapenem-resistant genes in enterobacteria from peruvian hospitals, 2013-2017. Rev Peru Med Exp Salud Publica 2018;35:259-64. https://doi.org/10.17843/rpmesp.25Y.25v25i.3474.
[7] Chicaiza-Taypanta JO. Enterobacterias multirresistentes en infecciones intrabdominales. Investigación clínica 2021;62:205.
[8] Rojas G, Vásquez Y, Rodríguez M, García P, Rojas-Faraco T. Mecanismos de resistencia a antibióticos betalactámicos en Enterobacterales aislados en hemocultivos, Maracay, estado Aragua, Venezuela. Kasmera 2021;49:e49235057. https://doi.org/10.5281/ZENODO.5377921.
[9] Spiess J, Fernández I, Gadea P, Romero S, Spiess C, Seija V, et al. Infecciones urinarias nosocomiales en un hospital universitario: INNOTEC 2022;7:4-15.
[10] Organización Mundial de la Salud. La OMS publica el primer informe mundial sobre prevención y control de infecciones (PCI). 2022.
[11] Cantón R, Cisneros JM. Tema 16. Infecciones por bacterias multirresistentes. En: Sociedad Española de Enfermedades Infecciosas y Microbiología Clínica, editor. Las enfermedades infecciosas en 2050: CÓMO SERÁN LAS ENFERMEDADES INFECCIOSAS EN 30 AÑOS, Producciones Pantuás; 2022.
[12] Sociedad Española de Enfermedades Infecciosas y Microbiología Clínica. Las enfermedades infecciosas en 2050. Producciones Pantuás; 2022.
[13] Organización Panamericana de la Salud, Organización Mundial de la Salud. Alerta Epidemiológica: Emergencia e incremento de nuevas combinaciones de carbapenemasas en Enterobacterales en Latinoamérica y el Caribe - 22 Octubre 2021, Washington, D.C. OPS/OMS. 2021.
[14] Yañez Alvarado EM, Ministerio de Salud, Centro Nacional de Epidemiología Prevención y Control de Enfermedades. BOLETÍN EPIDEMIOLÓGICO DEL PERÚ 2021. 2021.
[15] Worku M, Getie M, Moges F, Mehari AG. Extended-spectrum beta-lactamase- and carbapenemase-producing Enterobacteriaceae family of bacteria from diarrheal stool samples in Northwest Ethiopia. Interdiscip Perspect Infect Dis 2022;2022:7905350. https://doi.org/10.1155/2022/7905350.
[16] Sulaiman H, Roberts JA, Abdul-Aziz MH. Pharmacokinetics and pharmacodynamics of beta-lactam antibiotics in critically ill patients. Farm Hosp 2022;46:182-90. https://doi.org/10.7399/fh.13170.
[17] Rada AM, Hernández-Gómez C, Restrepo E, Villegas MV. Distribución y caracterización molecular de betalactamasas en bacterias Gram negativas en Colombia, 2001-2016. Biomedica 2019;39:199-220. https://doi.org/10.7705/biomedica.v39i3.4351.
[18] Gobernado M. Meropenem. Aspectos microbiológicos. Revista Española de Quimioterapia 2010;23:02-17.
[19] Arias Palacios J, Bustamante Ojeda S, Ortiz Gonzalez V, Moya Moreno M. Comparación de la actividad antimicrobiana de meropenem genérico y meropenem innovador por la técnica de micro dilución en cepas resistentes. Rev Cuba Farm 2015;49:0-0.
[20] Angles-Yanqui E, Huaringa-Marcelo J, Sacsaquispe-Contreras R, Pampa-Espinoza L. Panorama de las carbapenemasas en Perú. Rev Panam Salud Publica 2020;44:10.
[21] Ministerio de Salud. ALERTA EPIDEMIOLÓGICA: Riesgo de infecciones asociadas a la atención de la salud causadas por Enterobacterales, Pseudomonas aeruginosa y Acinetobacter spp. coproductoras de carbapenemasas en el Perú. 2022.
[22] Aguilar García CR, Martínez Torres C. La realidad de la Unidad de Cuidados Intensivos. Med Crit 2017;31:171-3.
[23] García Cruz CE, Zambrano Meza JA, Fajardo Herrera JM, Bowen Flores AE. Estudio Observacional: Uso de Antimicrobianos en la Unidad de Cuidados Intensivos del Hospital SOLCA Guayaquil-Ecuador: Publicaciones del Proyecto de Optimización de Uso de Antibióticos en Latinoamérica (PROA). Oncol (Guayaquil) 2019;29:165-78. https://doi.org/10.33821/450.
[24] Angarita Merchán M, Di Filippo Iriarte G, Mora Moreno DP, Ferrebuz Cardozo AJ. Perfil de resistencia de microorganismos circulantes en una Institución Prestadora de Servicios de salud en el Departamento de Boyacá, 2018. Rev investig salud Univ Boyacá 2019;6:120-44. https://doi.org/10.24267/23897325.327.
[25] Zurita Macalupú S, Ministerio de Salud, Instituto Nacional de Salud. Manual de Procedimientos de laboratorio: laboratorios locales I: laboratorios locales II. 2013.
[26] Ombelet S, Natale A, Ronat J-B, Vandenberg O, Hardy L, Jacobs J. Evaluation of MicroScan bacterial identification panels for low-resource settings. Diagnostics (Basel) 2021;11:349. https://doi.org/10.3390/diagnostics11020349.
[27] Clinical and Laboratory Standards Institute. M100. Performance Standards for Antimicrobial Susceptibility Testing. 33rd. 950 W Valley Rd Ste 2500, Wayne, Pennsylvania, 19087, United States: Clinical and Laboratory Standards Institute; 2023.
[28] Ali Abdel Rahim KA, Ali Mohamed AM. Prevalence of Extended Spectrum β-lactamase-Producing Klebsiella pneumoniae in Clinical Isolates. Jundishapur J Microbiol 2014;7:e17114. https://doi.org/10.5812/jjm.17114.
[29] Mayta-Barrios MM, Ramirez-Illescas JJ, Pampa-Espinoza L, Yagui-Moscoso MJA. Caracterización molecular de carbapenemasas en el Perú durante el 2019. Rev Peru Med Exp Salud Publica 2021;38:113-8. https://doi.org/10.17843/rpmesp.2021.381.5882.
[30] Muñoz C, Zumarán C, González T, Wozniak A, Castillo C, García P. Test evaluation and strategy proposal to detect and to characterize carbapenemase-producing gram negative bacilli. Rev Chilena Infectol 2017;34:326-32. https://doi.org/10.4067/s0716-10182017000400326.
[31] Instituto de Salud Carlos III (ISCIII). Detección fenotípica de enterobacterias productoras de carbapenemasas y pruebas de hidrólisis antibiótica (carbapenémico) e inmunocromatográficas. 02-2021.
[32] Fernández F, Martínez L, Pascual A, Perea EJ. Falsa resistencia a imipenema en bacilos gramnegativos mediante un sistema automatizado. Enferm Infecc Microbiol Clin 2000;18:500-5.
[33] Cantón R. Lectura interpretada del antibiograma: una necesidad clínica. Enferm Infecc Microbiol Clin 2010;28:375-85. https://doi.org/10.1016/j.eimc.2010.01.001.
[34] Garnacho-Montero J, Amaya-Villar R. El problema de la multi-resistencia en bacilos gram-negativos en las unidades de cuidados intensivos: estrategias de tratamiento y prevención. Med Intensiva 2022;46:326-35. https://doi.org/10.1016/j.medin.2021.12.002.
[35] Tranche Iparraguirre S, Martín Álvarez R, Párraga Martínez I, Junta Permanente y Directiva de la semFYC. El reto de la pandemia de la COVID-19 para la Atención Primaria. Rev clín med fam 2021;14:85-92. https://doi.org/10.55783/rcmf.140208.
[36] Bravo Ojeda JS. Descripción de tipos de carbapenemasas expresadas en Klebsiella sp. y Pseudomonas aeruginosa en hospitales de tercer nivel de la ciudad de Bogotá, estudio descriptivo. Parte 3: Comportamiento microbiológico y los mecanismos genéticos en aislamientos de Pseudomonas aeruginosa portadores del gen blaKPC en hospitales de tercer nivel de Bogotá. Universidad Nacional de Colombia, 2020.

[37]Duque Nossa VA, Varona Alzate A. Resistencia antimicrobiana asociada a dispositivos en UCI. Armenia-Quindío 2021 2022.
[38]Quintero Meza EV, Varón Rodríguez C. Caracterización de la colonización por Klebsiella pneumoniae resistentes a Carbapenémicos en pacientes provenientes de UCI. Universidad El Bosque Edu.co 2022. https://repositorio.unbosque.edu.co/bitstream/handle/20.500.12495/9132/MDRO%20pacientes%20UCI%20TESIS.pdf?sequence=1&isAllowed=y (accedido 28 de mayo de 2023).
[39]Vicente Castro MÁ. Bacterias aisladas con mayor frecuencia y perfil de resistencia antibiótica en cultivos y antibiogramas de muestras procedentes de la unidad de cuidados intensivos – Clínica Arequipa 2015 2016.
[40]Hernandez Bonifaz SG. Bacterias aisladas con mayor frecuencia en la unidad de cuidados intensivos del hospital regional de Ica - Perú 2019. Universidad Nacional San Luis Gonzaga, 2021.
[41]Malbrán G. C. Alerta Epidemiológica. Emergencia de Enterobacterales doble productores de Carbapenemasas; Instituto Nacional de Enfermedades Infecciosas (INEI), Ministerio Salud Argentina. 2021.
[42]Caicedo Loor S, Guerrero Loor JJ, Pachay Solórzano JW. Bacterias Gram negativas productoras de carbapenemasas aisladas de pacientes hospitalizados del Hospital General Portoviejo, Ecuador. QhaliKay Rev Cienc Salud 2023;7:43-50. https://doi.org/10.33936/qkrcs.v7i1.5221.
[43]Guerra Carías EI, Valenzuela Acevedo L, Velásquez Porta T. Caracterización de carbapenemasas en enterobacterias de muestras de pacientes que acudieron al Hospital General San Juan de Dios de la ciudad de Guatemala durante 2014 y 2015. Rev cient (Guatem) 2020.