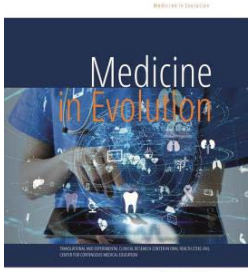
Carbapenemase-producing Enterobacteriaceae isolated in Arad Clinical Emergency Hospital in 2021



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Abstract

Knowing hospital circulation of antimicrobial - resistant bacteria is a must in present time, when "superbugs" are present in higher prevalence, in medical facilities and in community as well. In 2021, a pandemic year, hospitals were mainly dedicated to Covid 19 patients, with reduced activity for general population. Even so, data shows that Gram negative bacteria (GNB), multidrog resistant (MDR) and bacteria displaying carbapenemase-production are all higher at an alarming level. Our study demonstrate that MDR strains reached 52.67% for GNB and carbapemenase-producing Enterobacteriaceae were present in 24.24% for ESBLs, 10.05% for oxallicinases, 7.13% for KPC activity, 6.16% for OXA 48, 4.54% for MBLs and 1.70% for AmpC.

Keywords: Carbapenemase-producing Enterobacteriaceae, β-lactamases, AmpC, Metallo-β-lactamases (MBLs), Innovation, technology, research projects, etc

INTRODUCTION

The β-lactam family of antibiotic molecules consists of four groups: cephalosporins, monobactam, penicillins, and carbapenems [1].

Carbapenems are extremely effective in treating severe bacterial infections. This class of antibiotics is usually reserved for known or suspected multidrug-resistant (MDR) bacterial infections [2]. The spectrum of activity of the carbapenems imipenem, doripenem, and meropenem includes most Enterobacteriaceace species, including Escherichia coli, Klebsiella pneumoniae, Enterobacter cloacae, Citrobacter freundii, Proteus mirabilis, and Serratia marcescens; they are efficient against most strains of E. coli and K. pneumoniae resistant to cephalosporins due to the production of extended spectrum beta-lactamases. Imipenem, doripenem, and meropenem are active against most strains of Pseudomonas aeruginosa and Acinetobacter species [3].

Carbapenemase-producing Enterobacteriaceae CPE) are carbapenem-resistant Enterobacteriaceae (CRE) containing enzymes (e.g. OXA-48, KPC and so on) capable to break down the antibiotics and prevent them from killing the bacteria. Carbapenemases are carried on plasmids, and can easily transfer from one bacteria to another, resulting in an aggressive spread of these resistant pathogens. Pathogens harbouring these enzymes are sometimes called "superbugs", being generally resistant to most/all other antibiotics [4]. Various ESBLs (extended spectrum beta-lactamases) and carbapenemases have been reported in the Enterobacteriaceae including Enterobacter, Klebsiella, Escherichia coli [5], and other opportunistic species such as Serratia [6].

The Ambler Classification of β-lactamases, based on amino acid homology, is the most used method to classify β-lactamases, being considered the simplest classification scheme of β-lactamases [7] : Class A (TEM-1, 2, SHV-1ESBLs, KPC), Class B (MBLs, NDM, IMP, VIM), Class C (ampC, CMY), Class D (OXA).

Class A (KPC, ESBL), Klebsiella pneumoniae carbapenemase (KPC) - producing bacteria are a group of emerging highly drug-resistant Gram-negative bacteria. Although K. pneumoniae remains the most prevalent bacterial species carrying KPCs, the enzyme has been identified in several other Gram-negative bacilli. KPCs are an important mechanism of resistance for an increasingly wide range of Gram-negative bacteria and are no longer limited to K pneumoniae [8].

Class B (MBLs) Metallo-β-lactamases (MBLs) are transmissible carbapenemases of increasing prevalence in Gram-negative bacteria among health care facilities worldwide [9].

Class C (ampC), AmpC beta-lactamases are clinically important cephalosporinases encoded on the chromosomes of many of the Enterobacteriaceae and a few other organisms, where they mediate resistance to cephalothin, cefazolin, cefoxitin, most penicillins, and beta-lactamase inhibitor-beta-lactam combinations [10].

Class D (OXA) β-lactamases are characterized as penicillinases that can hydrolyze oxacillin and cloxacillin and are poorly inhibited by clavulanic acid and EDTA. OXA-48 is one of the few members of this family to possess notable carbapenem-hydrolyzing activity [11]. The OXA-48 enzyme is an Ambler class D beta-lactamase that hydrolyzes carbapenems but shows very weak activity against extended-spectrum cephalosporins such as cefepime and ceftazidime (third and fourth cephalosporine generation)

Objective. To assess prevalence of carbapenemase-producing enterobacteriaceae isolated in Arad Clinical Emergency Hospital in 2021.

MATERIAL AND METHODS

Retrospective prevalence study of enterobacteriaceae isolated in hospital´s microbiological laboratory, which were susceptibility to carbapemems (Antibacterial drug susceptibility was determined by the disk diffusion method and interpreted according to the European Committee on Antimicrobial Susceptibility Testing guidelines (www.eucast.org). Every isolate was analysed also using Ambler Classification system.

RESULTS

Of 2466 Gram negative bacteria, 1299 (52.67%) were MDR, mainly ESBLs (24.24%) and by carbapenemase oxacilinaze production (10.05), table 1. Percent of Carbapenemase-producing Enterobacteriaceae is displayed in table 2.

Table 1. Prevalence for MDR mechanisms in Gram negative bacteria in 2021

GNB carbapenemases	percent	Antimicrobial resistance
Class A-Beta-lactamase ESBL	24.24	Cefotaxime, ceftazidime, cefpodoxime aztreonam
Class D-OXA-23-like, OXA-24-like, OXA-48-like, OXA-58-like	10.05	Cephalosporins third and four generation, clavulanic acid, tazobactam and sulbactam, oxacillin and cloxacillin
Class A-Carbapenemase KPC activity	7.13	Aztreonam
Class D-Carbapenemase OXA 48	6.16	Cephalosporins third and four generation, clavulanic acid, tazobactam and sulbactam, oxacillin and cloxacillin
Class B -Carbapenemase MBLs	4.54	Aztreonam, ceftazidime, imipenem
Class C-Beta-lactamase AmpC	1.70	Aztreonam, cefoxitin

Legend: GNB=Gram negative bacteria, KPC= *Klebsiella pneumoniae* carbapenemase, OXA 48= carbapnemase OXA 48, MBLs= *Metallo-β-lactamases,* AmpC=cephalosporinases, OXA-23-like, OXA-24-like, OXA-48-like, OXA-58-like=carbapenemase oxacillinases

Enterobacteriaceae isolates (notably 224 *Klebsiella pneumoniae*, 19 *Escherichia coli*, 25 *Enterobacter cloacae*, 224 *Klebsiella pneumoniae,* 155 *Proteus penneri,*) were in total 2100 of which 34.76% (n= 730) displayed reduced susceptibility to carbapemems, table 2.

Table 2. Prevalence for carbapenems - resistant Enterobacteriaceae

Enterobacteriaceae	Frequency of resistant strains	total	Percent carbapenems-resistance
Citrobacter freundii	2	17	11.76
Enterobacter cloacae	25	87	28.74
Escherichia coli	19	875	2.17
Escherichia fergusonii	3	7	42.86
Klebsiella oxytoca	2	15	13.33
Klebsiella pneumoniae	224	418	53.59
Morganella morganii	7	46	15.22
Proteus mirabilis	2	5	40
Proteus penneri	155	256	60.55
Proteus rettgeri	14	17	82.35
Proteus vulgaris	6	16	37.5
Serratia liquefaciens	2	7	28.57
Serratia marcescens	5	31	16.13
Serratia odorífera	1	1	100
Total	730	2100	34.76

Table 3. Prevalence for MDR mechanisms in Carbapenemase-producing Enterobacteriaceae

Enterobacteriaceae	KPC activity	OXA 48	MBLs	oxacillinases	ESBLs	AmpC
Citrobacter freundii	0.00	0.00	0.00	0.00	5.88	0.00
Enterobacter aerogenes	0.00	0.00	0.00	0.00	13.33	0.00
Enterobacter cloacae	2.30	14.94	14.94	1.15	37.93	3.45
Escherichia coli	0.34	0.11	0.11	1.14	14.63	0.69
Klebsiella pneumoniae	22.73	20.33	20.33	1.67	58.13	1.67
Morganella morganii	4.35	2.17	2.17	0.00	34.78	4.35
Proteus penneri	7.42	16.41	16.41	0.78	53.52	7.03
Proteus rettgeri	5.88	17.65	17.65	5.88	70.59	0.00
Proteus vulgaris	0.00	18.75	18.75	0.00	18.75	0.00
Serratia marcescens	6.45	0.00	0.00	0.00	54.84	3.23

Legend: GNB=Gram negative bacteria, KPC= Klebsiella pneumoniae carbapenemase, OXA 48= carbapnemase OXA 48, MBLs= Metallo-β-lactamases, AmpC=cephalosporinases, OXA-23-like, OXA-24-like, OXA-48-like, OXA-58-like=carbapenemase oxacillinases

Circulation of MDR bacteria was frequent mainly in intensive care and palliative departments, (p=0.000) figure 1.

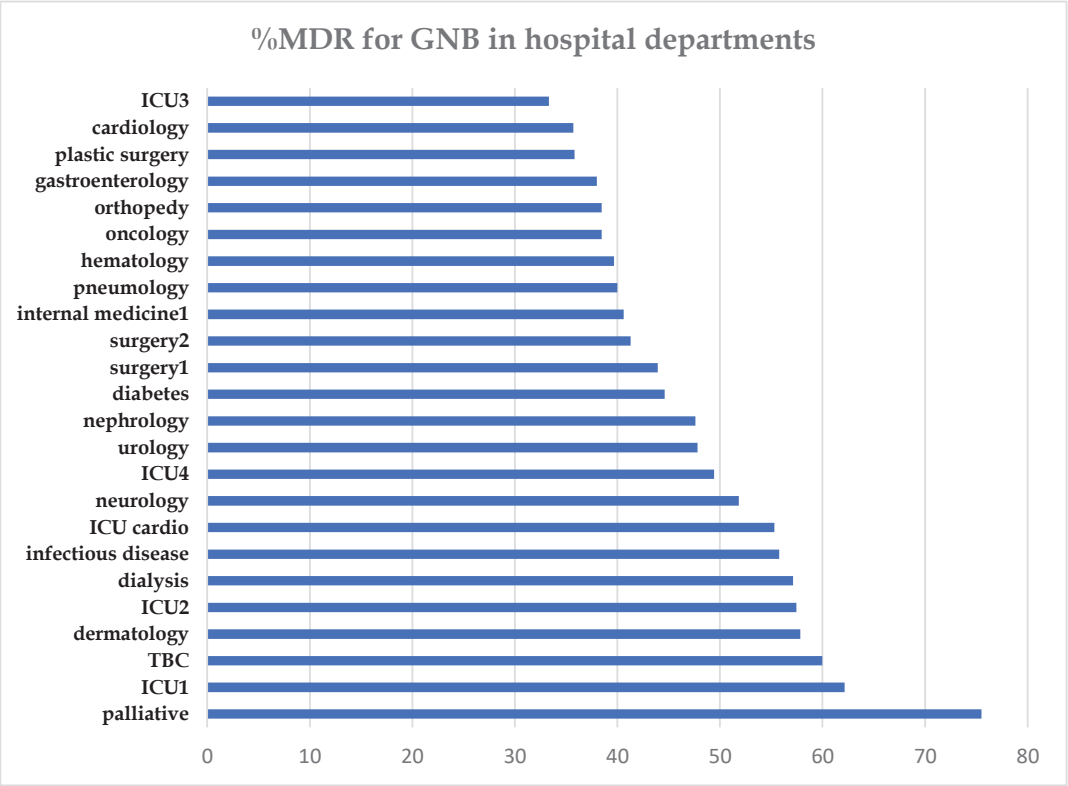

Figure 1. Department distribution for MDR GNB

Legend: ICU1= intensive care unit 1, ICU2= intensive care unit 2, ICU3= intensive care unit 3, ICU4= intensive care unit 4

DISCUSSIONS

Carbapenemase-producing Enterobacteriaceae were almost unknown up to the 1990s, today being encountered routinely in almost all hospitals and other healthcare facilities in many countries. KPC-producing *Klebsiella pneumoniae* was the first to emerge and spread

globally and is endemic in the United States, Israel, Greece, and Italy. Recently, NDM-producing Enterobacteriaceae and OXA-48-producing K. pneumoniae appear to be disseminating from South Asia and Northern Africa, respectively. They are almost always resistant to all β-lactams including carbapenems and many other classes. [12].

The results of this study, where most isolates were identified in the palliative center, overlap with the undesirable results of other countries, for instance The Centre for Health Protection of the Department of Hong Kong Health investigated in December 21 last year a cluster of Carbapenemase-producing Enterobacteriaceae at a residential care home for the elderly in Sham Shui Po, and reminded the public on maintaining strict personal and environmental hygiene, and proper use of antibiotics[13]. More, Ireland Health Protection Surveillance Centre has a complete document on Carbapenemase-producing Enterobacteriaceae in palliative care, intended for healthcare professionals working in palliative care [13].

CONCLUSIONS

The prevalence of the β- lactamases in the hospital isolates emphasizes the need for an insistent surveillance of resistant strains, guidelines for the antibiotic therapy and the implementation of infection control measures to reduce the increasing burden of antibiotic resistance. Mortality from invasive Carbapenemase-producing Enterobacteriaceae infections reaches up to 40%. To obtain the maximal benefit from the limited options available, dosing of antimicrobial agents should be optimized based on pharmacokinetic data, especially for colistin and carbapenems. In addition, multiple observational studies have associated combination antimicrobial therapy with lower mortality compared with monotherapy for these infections. The outcomes appear to be especially favorable when patients are treated with a carbapenem and a second agent such as colistin, tigecycline, and gentamicin, but the best approach is yet to be defined. [12].

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