Review / *Derleme*

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Carbapenemases

Karbapenemazlar

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Abstract

AIn recent years, infections due to multidrug-resistant gram-negative bacilli (MDR-GNB) are increasing worlwide. Carbapenem hydrolyzing enzymes (also known as carbapenemases) are responsible of carbapenem resistance which are mostly used in MDR-GNB infections. This resistance mechanism is different from other mechanisms of carbapenem resistance such as activation of multidrug efflux pumps in response to antibiotic exposure and impaired permeability due to porin mutations. However, all types of resistance mechanisms lead MDR-GNB infections MDR-GNB infections are associated with longer hospital stays and life-threatening severe infections. In this review, we aimed to present current data of carbapenemases.

Keywords: Carbapenem hydrolyzing enzymes, multidrug-resistant gram-negative bacilli

Özet

Son yıllarda bütün dünyada çoğul ilaca dirençli gram-negatif çomaklar (ÇİD-GNÇ) ile gelişen enfeksiyonların görülme sıklığı artmaktadır. Karbapenemazlar olarak da bilinen karbapenem yıkıcı enzimler, ÇİD-GNÇ ile gelişen enfeksiyonların tedavisinde kullanılan karbapenem kümesi antibiyotiklere karşı direnç gelişiminden sorumludur. Bu direnç düzeneği, antibiyotikle karşılaşmaya yanıt olarak çoğul ilaç dışatım pompalarının etkinleşmesi ve porin mutasyonu sonucu gelişen bozulmuş geçirgenlikten farklıdır. Ancak bütün direnç düzenekleri ÇİD-GNÇ ile gelişen enfeksiyonlara yol açmaktadır. Bu enfeksiyonlar, hastane yatış süresinin uzaması ve yaşamı tehdit eden önemli hastalık durumlarıyla ilişkilidir. Bu derlemede karbapenemazlara ilişkin güncel bilgilerin sunulması amaçlanmıştır.

Anahtar Kelimeler: Karbapenem yıkıcı enzimler, dirençli gram-negatif enfeksiyonlar

Introduction

Multidrug-resistant gram-negative bacilli (MDR-GNB) are responsible for more than half of the infections related with healthcare. MDR-GNB infections lead to prolonged hospital stays, increase in hospital expenses and in mortality rates (1). Carbapenemases are vital antibiotic clusters that can show efficiency against chromosome-induced cephalosporinases with many gram-negative agents and against extended spectrum beta-lactamases (ESBL) and are used in the treatment of infections developing with these resistant agents. Prevalent carbapenem use in recent years in the treatment of agent infections producing ESBL and AmpC type beta-lactamases has also resulted in resistance to these antibiotics. The following is a summary of the primary resistance mechanisms that develop against carbapenems: (i) reduced membrane cut-off due to porin loss; (ii) activation of efflux pump enabling antibiotic excretion (iii) occurrence of carbapenemases (carbapenem-destructive enzymes). All of these reasons have given rise to the limitation of the use of these antibiotics and to the emergence of the definition of carbapenem-resistant gram-negative bacilli (CR-GNB) (2,3).

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Table 1. Carbapenemase classifications

	Carbapenemase	Agent it is most commonly found in	Gene placement
Class A Carbapenemases	SME-1, SME-2, SME-3	*S. marcescens*	Chromosome-derived
	IMI-1, IMI-2	*E. cloacae*	Chromosome-derived
	NMC	*E. cloacae*	Chromosome-derived
	KPC-1, KPC-2, KPC3, KPC-4	*K. pneumoniae, K. oxytoca, Enterobacter* spp., *E. coli, P. aeruginosa, C. freundii*	Plasmid
	GES-2, GES-4, GES-5,GES-6	*P. aeruginosa, K. pneumoniae, E. coli*	Plasmid
Class B Carbapenemases	IMP (Imipenemases)	*P. aeruginosa, A. baumannii, Enterobacteriaceae*	Plasmid
	VIM (Verona integro metalo-β-lactamase)	*P. aeruginosa, Enterobacteriaceae*	Plasmid
	SPM-1 (Sao Paulo metalo-β-lactamases)	*P. aeruginosa*	Plasmid
	GIM (German imipenemase)	*P. aeruginosa, S. marcescens, E. cloacae*	Plasmid
	SIM-1 (Seoul imipenemase)	*P. aeruginosa, A. baumannii*	Plasmid
	NDM (New Delhi metalo-β-lactamase)	*Enterobacteriaceae, A. baumannii*	Plasmid
Class D Carbapenemases (the most commonly seen)	OXA-51	*A. baumannii*	Chromosome-derived
	OXA-23,OXA-24/OXA40, OXA-48, OXA-58	*A. baumannii, Enterobacteriaceae*	Plasmid

Carbapenemases were defined for the first time in the 1990s for the agents that are in and not in the Enterobacteriaceae family and have become a great risk element in the world due to their quick spread in the present day (4,5). Carbapenemases are divided into four classes according to Abler molecular classification. While Class A, C, and D carbapenemases contain the amino acid serine in their effective parts, Class B carbapenemases necessitate zinc for efficiency, and therefore, are referred to as metallo-beta-lactamase (MBL). Agents producing A, B and D class carbapenemases are essentially responsible for healthcare-related infections (6).

Class A carbapenemases: Chromosomally encoded (chromosome-derived) SME (Serratia marcescens enzyme), NMC (non-metallo-enzyme carbapenemase) and IMI (imipenem destructive, hydrolyzing beta-lactamase) enzymes are contained. KPC (Klebsiella pneumoniae carbapenemase) and GES (Guiana extended spectrum) are present in primary enzymes encoded through plasmids.

Class B carbapenemases (metallo-beta-lactamase): Just as there are naturally-produced MBLs, there are enzymes adapted through plasmids between species. IMP, VIM, GIM, SPM, SIM and NDM-1 (New Delhi metallo-beta-lactamae-1) enzymes belong to this class.

Class D carbapenemases: Known as OXA-type enzymes for principally breaking down oxacillin. Together with containing more than 100 enzymes in the OXA cluster, the most commonly known are chromosomally encoded OXA-51 and OXA-23, OXA-24/OXA40, OXA-48, and OXA-58 that can be carried with plasmids (7).

In addition to the widely used molecular classification, carbapenemases are also classified functionally in accordance with characteristics like target destruction and prevention. Carbapenemases, as regards Karen Bush classification, are divided as 2f, 2d and 3 clusters (8).

The Distribution of Carbapenemases

Klebsiella pneumoniae carbapenemase (KPC) found in Class A has been firstly detected in the United States of America (USA) and Class B metallo-beta-lactamases in the Far East (9,10). Even though Class D OXA-type beta-lactamases constitute a problem for especially Europe, it can be said for today that all carbapenemases have been identified in many parts of the world.

K. pneumoniae carbapenemase (KPC) is the most frequently encountered one in the USA. It was identified for the first time in the 1990s in North Carolina and reported to have been detected in 36 states in 2010 (11,12). KPC has been confirmed in many countries in Europe, Asia and South America. MBLs were obser-

Table 2. Target destruction and prevention characteristics of carbapenemases

Molecular class	Functional cluster	Enzyme	Target destruction					Prevention	
			Penicillin	1. GC	ESC	Aztreonam	Carbapenem	EDTA	Clavulonic acid
A	2f	NMC	+	+	+	+	+	-	+
		SME	+	+	±	+	+	-	+
		KPC	+	+	+	+	+	-	+
		IMI	+	+	+	+	+	-	+
		GES	+	+	+	-	±	-	+
B1	3	IMP	+	+	+	-	+	+	-
		SPM	+	+	+	-	+	+	-
		GIM	+	+	+	-	+	+	-
		VIM	+	+	+	-	+	+	-
D	2d	OXA	+	+	±	-	±	-	±

1. GC: 1. Generation cephalosporin; ESC: Extended-spectrum cephalosporin.

ved for the first time in Japan in 1991, and thereafter, detected in Taiwan, Singapore, Korea, Hong Kong, China, Brazil and Columbia and identified in many countries in Europe and in the USA (10). NDM-1, which has increased its incidence lately, was firstly detected in a Swedish patient admitted to a hospital in India in 2009. NDM-1 has spread quickly afterwards and determined in many places in Asia, Europe, North America, and Australia (13). Specifically seen in Europe, oxacillinases are observed in Asia, Australia and the USA.

2013 data of European Center for Disease Prevention and Control (ECDC) and 2015 data of World Health Organization show that the data of our country regarding agents producing carbapenemases is limited. The first OXA-48 enzyme in *K. pneumoniae* was detected in our country in 2003 (14). The first *K.pneumoniae* outbreak resistant to carbapenem carrying OXA-48 enzyme was reported from Istanbul in 2008 (15). According to studies carried out in adults in our country, OXA-48 is the most frequently found carbapenemase of the *Enterobacteriaceae* family and IMP and NDM-1 enzymes have also been confirmed in *K.pneumoniaea* (16,17). The most commonly isolated resistance enzyme in A, baumanii isolates is OXA-23, which is followed respectively by OXA-58, OXA-24 and GES (18-20). Iraz et al. have detected VIM and GES enzymes in the *P. aeruginosa* isolate resistant to 23 carbapenems (21). Studies on pediatric patients in our country are even more limited. NDM-1 has been encountered in Enterobacter cloacae in 8 patients admitted in the pediatric intensive care unit in Istanbul (22). In one of our studies including 176 pediatric cases colonized with CR-GNB, most frequently OXA-48 in *Enterobacteriacea,* GES in pseudomonases, and NDM and OXA-23 in *A. baumanii* have been isolated. Moreover, NDM enzyme in *A. baumanii* has been detected for the first time in Turkey in this study (23).

It was reported in the 2015 data of European Center for Disease Prevention and Control that carbapenem-resistant Enterobacteriacea (CRE) gradually became widespread. While inter-regional spread or endemic state in a total of 6 countries was detected in the 2013 data, this condition was determined in 13 countries in the 2015 data. While NDM was reported to be seen in hospital outbreaks one by one in Italy and England in the 2013 data, single hospital outbreaks in 5 countries and inter-regional spread in 7 countries were established in the 2015 data. Similarly, OXA-48 has shown fast spread and while it was confirmed to be endemic in 1 country in 2013, it was found to have shown endemic state in two countries and inter-regional spread in 4 in the 2015 data (25).

Risk Factors

Carbapenem use in the colonization or infection development with carbapenem-resistant gram-negative bacilli (CR-GNB) has been demonstrated to be a leading risk factor (26). Presence of an underlying disease (diabetes mellitus, malignant disease, organ donation), admission to intensive care unit, being under mechanical ventilation, nasogastric tube use, prolonged hospital stay, urinary tract and venous catheter use are among the known risk factors (27,28).

Ma et al. have found carbapenem use of more than 4 days to be a risk factor for colonization with CR-GNB (29). Kumar et al. have indicated prolonged anti-microbial drug use and long-term ventilation necessity as risk factors (30). Logan et al. have specified history of antibiotic use, prolonged hospital stay and use of invasive equipment to be risk factors in their evaluation of studies conducted on pediatric patients (31).

Studies on pediatric patients conducted in our country are quite limited. Ulu-Kılıç et al. designated carbapenem use and prior hospital admission and stay to be risk factors in 83 patients

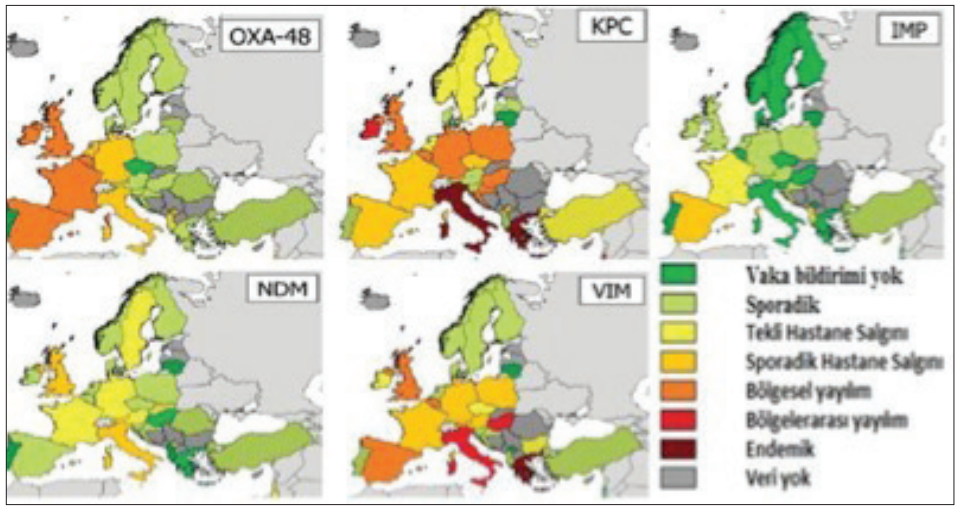

Figure 1. Distributions of carbapenemases in carbapenem-resistant *Enterobacteriacea* in European countries (taken from the 2013 data of European Center for Disease Prevention and Control) (24)

colonized with carbapenem-resistant *K. pneumoniae* in 2012 (32). In one study of ours, we have established carbapenem use, ampicillin use, being under the age of 1, nasogastric tube use, underlying chronic disease and prior surgical procedure to be independent risk factors for colonization in 176 patients colonized with CR-GNB (23).

Identification

As a result of the amendments made by Clinical and Laboratory Standards Institute (CLSI) in sensitivity values in 2010, sensitivity limit value was determined as ≤ 0.5 µg/mL for ertapenem and as ≤ 1 µg/mL for imipenem, meropenem and doripenem (33). In the 2013 data of EUCAST (European Committee on Antimicrobial Susceptibility Testing), sensitivity limit value was put forth as ≤ 2 mg/L for imipenem and meropenem (34). Modified Hodge Test (MHT) is a phenotypical examination that can be used as a carbapenemase verification test, but not sufficient enough on its own. Combination disc method is a phenotypical test that has superiorities like being easily found commercially. Polymerase chain reaction (PCR) is a genotypic, expensive test used in identification in reference laboratories (35).

Clinic

Agents producing carbapenemase can cause clinical infections such as asymptomatic colonization or blood flow, catheter-related, ventilation-related, urinary tract and wound site infections. There are not many studies regarding how many of the colonized patients will develop infections. Thus, foreseeing infection development is not easy beforehand.

Borrer et al. and Wiener-Weil et al. have detected infection development rate in patients colonized with CR-*Enterobacteriacea* as 9% and 5%, respectively (28,36). While Jung et al. have indicated an infection rate of as high as 54% in patients colonized with *A. baumannii* (37), infection did not develop in patients colonized with *P. aeruginosa* in two studies (38,39).

Ulu-Kılıç et al. have indicated an infection development rate of 3.4% in colonized pediatric patients, and clinical diseases respectively comprised bacteremia, urinary tract infection and soft tissue infection (32). Infection rate, in one of our studies, was detected similarly as 3.4% and bacteremia and urinary tract infection developed as clinical infections (23).

Treatment

The most favorable treatment of infections developing with agents producing carbapenemase is not absolute. Aztreonam and phosphomycine, individually, can be an option in the treatment of infections with CR-GRB; however, their use is limited due to the fact that there are forms that can be used via venous blood vessel, which is problematic in some regions, there are not sufficient data regarding Aztreonam, and the fact that phosphomycine has limited efficiency in the treatment of urinary tract infections. At the heart of the study, the use of meropenem with the treatment of prolonged infusion, tigecycline use, combination treatment of rifampin or aminoglycosides have been reported in the treatment of infections developing with CR-GNB (40,41). Today, as a result of many studies, combination treatments are recommended in the treatment of infections developing with CR-GNB. The aim in giving combination treatment is to extend anti-microbial development, benefit from the synergy effect shown in vitro and prevent resistance that can develop during treatment. Synergy effect means twofold germicidal effect by using two drugs together instead of one and is vital in the treatment of CR-GNB (42,43). In our study, infection developed in 6 out of the 176 patients colonized with CR-GNB; 4 of these patients were treated successfully with cholistin and meropenem com-

bination, and two with urinary tract infection were treated with ciprofloxacin according to antibiotic sensitivity results (23).

In a multicenter study conducted in Italy with 125 CR-GNB patients, mortality rate has been detected as 54% in patients receiving single drug, 30% in patients receiving two drugs (cholistin+tigecycline) and 12.5% in patients receiving three drug combination (tigecycline + cholitin+meropenem) (44). In a study by Qureshi et al. conducted in two centers in the USA, while mortality rate has been found as 66% in single-drug treatments with either cholistin or tigecycline, this rate dropped to 12% when either has been used with carbapenem (45). In a study from India where 42 patients that developed blood flow infection with CR-GNB and different treatments were compared, treatment was found to be 100% successful with the combined use of cholistin and carbapenem (46).

Owing to the fact that tigecycline use in children is very limited, cholistin is the most important drug in use. It can be stated that giving cholistin with combination treatment is the primary option in infections developing with CR-GNB.

Inspection Measures

Contact isolation must be implemented to infected patients or patients colonized with CR-GNB. Reasonable antibiotic use, attention to the use of invasive equipment and standard measures like hand washing must be cared. Just as in our study, clustering can be made by rectum colonization scanning in the event of observing patients with CR-GNB infection, and disease spreading can be avoided (23).

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