

Rapid Detection of Carbapenemase Producing Bacterial Isolates

Thesis

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List of Contents

Title	Page No.
List of Abbreviations	i
List of Tables	iv
List of Figures.....	vi
Introduction	1
Aim of the Work	4
Review of Literature	
▪ Carbapenem Antibiotics.....	5
▪ Mechanism of Carbapenem Resistance	14
▪ Laboratory Detection of Carbapenemase	36
Materials and Methods	63
Results	90
Master Table	105
Discussion.....	108
Conclusion and Recommendations.....	118
Summary	120
References	123
Arabic Summary	

List of Abbreviations

Abb.	Full term
A. baumannii	Acinetobacter baumannii
AIM	Australian Imipenemase
APBA	3-AminoPhenyl Boronic Acid
aPI	Acute Pelvic Infections
BA	Boronic Acid
CAP	Community Acquired Pneumonia
CHDLs	Carbapenem Hydrolyzing class D β -Lactamases
cIAI	Complicated Intra-Abdominal Infections
CLSI	Clinical and Laboratory Standards Institute
CRE	Carbapenem Resistant Enterobacteriaceae
cSSSI	Complicated Skin and Skin Structure Infections
cUTI	Complicated Urinary Tract Infection
DIM	Dutch Imipenemase
E. asburiae	Enterobacter asburiae
E. coli	Escherichia coli
E. faecium	Enterococcus faecium
E.cloacae	Enterobacter cloacae
E.faecalis	Enterococcus faecalis
EDTA	Ethylene Diamine Tetra-acetic Acid
ESBL	Extended Spectrum β -Lactamase
EUCAST	European Committee on Antimicrobial Susceptibility Testing
FDA	Food and Drug Administration

List of Abbreviations (Cont...)

Abb.	Full term
GES	Guiana Extended Spectrum
GIM	German Imipenemase
GNB	Gram Negative Bacilli
H.influenzae	Haemophilus influenzae
HAP	Hospital Acquired Pneumonia
HMM	High Molecular Mass
HS	Highly Significant
IEF	Isoelectric Focusing
IMI	Imipenem hydrolyzing β -lactamase
IMP	Active on Imipenem
K.pneumoniae	Klebsiella pneumoniae
KHM	Kyorin Health Science Metallo- β -lactamase
KPC	Klebsiella Pneumoniae Carbapenemase
LAMP	Loop Mediated Isothermal Amplification
LMM	Low Molecular Mass
MALDI-TOF	MS Matrix Assisted Laser Desorption Ionization Time of Flight Mass Spectrometry
MBLs	Metallo- β -Lactamases
MHT	Modified Hodge Test
MICs	Minimal Inhibitory Concentrations
MRSA	Methicillin Resistant Staphylococcus Aureus
MSSA	Methicillin Sensitive Staphylococcus Aureus
NDM	New Delhi Metallo- β -lactamase

List of Abbreviations (Cont...)

Abb.	Full term
NGS	Next Generation Sequencing
NMC	Non Metalloenzyme Carbapenemase
NS	Non Significant
NPV	Negative Predictive Value
OMPs	Outer Membrane Proteins
OXA	Oxacillinases
P. aeruginosa	Pseudomonas aeruginosa
PAE-MHT	Pseudomonas Aeruginosa Modified Hodge Test
PBPs	Penicillin Binding Proteins
PCR	Polymerase Chain Reaction
PPV	Positive Predictive Value
RTI	Respiratory Tract Infections
S. maltophilia	Stenotrophomonas maltophilia
S.marcescens	Serratia marcescens
S.pneumoniae	Streptococcus pneumoniae
S.pyogenes	Streptococcus pyogenes
SIM	Seoul Imipenemase
SME	Serratia Marcescens Enzyme
SPM	Sao Paulo Metallo- β -lactamase
S	Significant
SSI	Surgical Site Infection
VAP	Ventilator Associated Pneumonia
VIM	Verona Integron encoded Metallo- β -lactamase
WGS	Whole Genome Sequencing

List of Tables

Table No.	Title	Page No.
Table (1):	Comparison of carbapenem antibiotics	11
Table (2):	Novel carbapenems.....	13
Table (3):	Substrate and inhibition profiles of the carbapenemases	17
Table (4):	Class A carbapenemases.....	20
Table (5):	KPC-variant carbapenemases	24
Table (6):	Metallo- β -lactamases.....	30
Table (7):	Carbapenem-hydrolyzing class D β -lactamases.....	32
Table (8):	EUCAST 2015 and CLSI 2015 guidelines for zone diameter by disc diffusion method	37
Table (9):	EUCAST 2015 and CLSI 2015 MIC guidelines	39
Table (10):	Interpretation of BA-MHT, OXA-MHT	47
Table (11):	Components of reaction master mix for each 25 μ l reaction for each primer.....	80
Table (12):	Three-step cycling protocol.....	81
Table (13):	Different types of isolates	90
Table (14):	Results of Carba NP test in <i>Enterobacteriaceae</i>	91
Table (15):	Results of Carba NP test in <i>Pseudomonas</i>	92
Table (16):	Results of CarbAcineto NP test	93
Table (17):	Results of CHROMagar KPC	93
Table (18):	Distribution of <i>bla</i> _{KPC} among <i>Acinetobacter</i> , <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp.....	94
Table (19):	Distribution of <i>bla</i> _{VIM} among <i>Acinetobacter</i> , <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp.....	95

List of Tables (cont...)

Table No.	Title	Page No.
Table (20):	Distribution of <i>bla</i> _{OXA} gene among <i>Acinetobacter</i> , <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp.....	96
Table (21):	Distribution of <i>bla</i> _{GES} gene among <i>Acinetobacter</i> , <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp.....	97
Table (22):	Distribution of <i>bla</i> _{IMP} gene among <i>Acinetobacter</i> , <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp.....	98
Table (23):	Distribution of the tested genes among <i>Acinetobacter</i> , <i>Enterobacteriaceae</i> and <i>Pseudomonas</i> spp.....	99
Table (24):	Correlation between Carba NP and PCR results in <i>Enterobacteriaceae</i>	100
Table (25):	Correlation between Carba NP and PCR results in <i>Pseudomonas</i>	101
Table (26):	Correlation between CarbAcineto NP and PCR results in <i>Acinetobacter</i>	102
Table (27):	Results of CHROMagar KPC in relation to PCR results.....	103
Table (28):	Diagnostic performance of Carba NP test, CarbAcineto NP test and CHROMagar KPC compared to PCR.....	103
Table (29):	Results of Carba NP and CarbAcineto NP tests in relation to PCR in <i>Enterobacteriaceae</i> , <i>Pseudomonas</i> and <i>Acinetobacter</i> spp. and its diagnostic performance compared to PCR.....	104

List of Figures

Fig. No.	Title	Page No.
Figure (1):	Chemical structures of (a) imipenem; (b) meropenem; (c) biapenem; (d) ertapenem; and doripenem.....	6
Figure (2):	Mechanism of carbapenem resistance	15
Figure (3):	CHROMagar KPC (carbapenemase producers)	41
Figure (4):	Features of colonies growing on Brilliance CRE Agar.....	42
Figure (5):	Modified Hodge test using a 10µg ertapenem disc.....	45
Figure (6):	Interpretation of the combination meropenem disc test.....	49
Figure (7):	Double disk synergy test.....	50
Figure (8):	E-test showing Carbapenemase production	51
Figure (9):	Microtitration plate showing the results of the Carba NP test	54
Figure (10):	Representative results obtained using the Rapidec Carba NP test on three isolates.....	54
Figure (11):	Carba NP test	73
Figure (12):	Metallic blue colonies of <i>Klebsiella</i> , <i>Citrobacter</i> or <i>Enterobacter</i>	75
Figure (13):	Dark pink to red colonies of <i>E.coli</i>	75
Figure (14):	Translucent cream colonies of <i>Pseudomonas</i>	76
Figure (15):	Opaque cream colonies of <i>Acinetobacter</i>	76
Figure (16):	Real-time multiplex PCR melting curves of the amplicons generated by primers targeting four carbapenemase types.....	82
Figure (17):	A positive isolate for <i>bla</i> _{KPC} , C _t = 13.....	82

List of Figures (Cont...)

Fig. No.	Title	Page No.
Figure (18):	A negative isolate for <i>bla</i> _{KPC}	83
Figure (19):	A positive isolate for <i>bla</i> _{VIM} , C _t = 32.....	83
Figure (20):	A negative isolate for <i>bla</i> _{VIM}	84
Figure (21):	A positive isolate for <i>bla</i> _{OXA-48} , C _t = 32.....	84
Figure (22):	A negative isolate for <i>bla</i> _{OXA-48}	85
Figure (23):	A positive isolate for <i>bla</i> _{GES} , C _t = 30.....	85
Figure (24):	A negative isolate for <i>bla</i> _{GES}	86
Figure (25):	A positive isolate for <i>bla</i> _{IMP} , C _t = 35.....	86
Figure (26):	A negative isolate for <i>bla</i> _{IMP}	87
Figure (27):	Different types of isolates.....	90
Figure (28):	Results of Carba NP test in <i>Enterobacteriaceae</i>	91
Figure (29):	Results of Carba NP test in <i>Pseudomonas</i>	92
Figure (30):	Results of CarbAcineto NP test.....	93
Figure (31):	Distribution of <i>bla</i> _{KPC} among different isolates.....	94
Figure (32):	Distribution of <i>bla</i> _{VIM} among different isolates.....	95
Figure (33):	Distribution of <i>bla</i> _{OXA} among different isolates.....	96
Figure (34):	Distribution of <i>bla</i> _{GES} among different isolates.....	97
Figure (35):	Distribution of <i>bla</i> _{IMP} among different isolates.....	98
Figure (36):	Distribution of the tested genes among different isolates.....	99
Figure (37):	Correlation between Carba NP test and PCR results in <i>Enterbacteriaceae</i>	100

List of Figures (Cont...)

Fig. No.	Title	Page No.
Figure (38):	Correlation between Carba NP test and PCR results in <i>Pseudomonas</i>	101
Figure (39):	Correlation between CarbAcineto NP test and PCR results in <i>Acinetobacter</i>	102

Abstract

Carba NP and CarbAcineto NP tests are highly sensitive and specific, cheap, rapid tests for the detection of carbapenem resistance due to carbapenemase production. So implementation of infection control measures can be achieved to prevent their further dissemination.

CHROMagar KPC is an easy screening method for the detection of carbapenem resistance, however it lacks specificity in detection of carbapenemase production.

Although PCR is a rapid and highly sensitive method in detection of Carbapenemase production, it is very expensive and needs trained personnel.

Key words: High Molecular Mass- Hospital Acquired Pneumonia- Haemophilus influenzae- Klebsiella pneumoniae- Kyorin Health Science Metallo- β -lactamase- Low Molecular Mass- Modified Hodge Test- Metallo- β -Lactamases.

INTRODUCTION

Carbapenems are commonly considered as last-resort antibiotics in the treatment of severe infections caused by multidrug-resistant gram negative bacteria. Over the past decade, the emergence of carbapenem-resistant isolates is very unnerving, since it contributes to the reduction of the current therapeutic pallet, therefore leaving few or, in some cases, no optimal therapeutic options (*Voulgari et al., 2013*).

Increased carbapenem MICs in gram-negative bacteria can be a result of two different mechanisms of resistance: hyperproduction of class C β -lactamases or extended- spectrum β -lactamases (ESBLs) in combination with porin alteration; and/or carbapenemase production (*Monterio et al., 2012*). Although several mechanisms of carbapenem resistance have been reported, most of the mechanisms are related to the spread of carbapenemases belonging to Ambler class A (KPCs), class B (VIMs, IMPs, and NDM-1), and class D (OXA-48) β -lactamases (*Nass et al., 2011*).

Klebsiella pneumonia carbapenemases (KPCs) have been found in bacteria other than *Klebsiella pneumoniae*, including *Klebsiella oxytoca*, *Proteus mirabilis*, *Acinetobacter* spp, *Pseudomonas aeruginosa*, *Citrobacter freundii*, *Serratia marcescens* and *Escherichia coli*. Bacterial isolates producing KPCs are able to hydrolyze a broad spectrum of β -lactams including the penicillins, cephalosporins, carbapenems and

monobactam. They have the potential to spread rapidly in hospital environments to cause nosocomial infections with high mortality rates (*Wang et al., 2012*).

Rapid and accurate detection of carbapenemase-producing strains is pivotal for adequate antibiotic therapy and infection control, especially in an outbreak setting (*Stuart et al., 2012*).

The detection of carbapenemases primarily on the basis of phenotypic testing may sometimes be difficult, since the MICs of carbapenem drugs may remain in the susceptibility range. Detection and identification of these carbapenem-resistant isolates may be improved significantly by the use of automated susceptibility testing systems (*Nordmann et al., 2011*).

A series of non-molecular-based tests (e.g. modified Hodge test (MHT) and inhibition studies by EDTA and dipicolinic acid) have been proposed for detection of carbapenemase activity. None of the currently available tests has 100% specificity or 100% sensitivity (*Nordmann et al., 2012b*).

A variety of culture techniques have been proposed for screening of carbapenem resistant isolates including both in-house-prepared selective media in broth or solid agar form (*Calfee and Jenkins, 2008*) and commercially available agar plates either chromogenic or non- chromogenic media (*Virioni et al., 2012*).