Indian Journal of Basic and Applied Medical Research; June 2020: Vol.-9, Issue- 3, P. 103 -113
DOI: 10.36848/IJBAMR/2020/12225.51565

Review article

Carbapenam resistance in *Klebsiella Pneumoniae*: An Overview.

[1]Praful S. Patil*, [2]Dr. Harshada Shah, [3]Dr. Smita Damke , [4]Dr. M. S. Chandel

[1]Ph.D Scholar, Index Medical College, Malwanchal University, Indore.

[2]Professor and Head, Department of Microbiology, Index Medical College, Malwanchal University, Indore.

[3]Associate Professor, Department of Microbiology, Jawaharlal Medical College, Wardha

[4]Assistant registrar, Academic Malwanchal University, Indore.

Corresponding author: Praful S. Patil*, Ph.D Scholar, Index Medical College , Malwanchal University, Indore. Email id: prafulp036@gmail.com

Abstract

In recent years, antibiotic resistance has become a serious threat to public health as bacteria are increasingly developing resistance to antibiotics while drug manufacturing companies are not developing new antibiotics that can compact the menace of drug resistance. Carbapenems are among the most effective antibiotics prescribed for treatment of life threatening infections caused by gram negative bacteria including *Enterobacteriaceae*. However in recent times, carbapenem resistance in *Enterobacteriaceae* is an ongoing public-health globally. *Klebsiella pneumoniae* is one of the most common isolate from *Enterobacteriaceae*. It causes variety of infections including health care associated infections. In recent years, *K. pneumoniae* has developed resistance to multiple antibiotics including extended spectrum β-lactam and carbapenem antimicrobial agents. In this present review article, the overview of carbapenam resistance in *K. Pneumoniae* is presented with emphasis on historical perspectives, epidemiology, laboratory identification and prevention and control.

Keywords: Carbapenems, carbapenemases, carbapenam resistance *Enterobacteriaceae, Klebsiella pneumoniae*.

Introduction:

Even in the golden era of medicine, where diagnostic and therapeutic modalities have progressed by leaps and bounds, infectious diseases still continue to be one of the major causes of morbidity and mortality in clinical setups worldwide. Infectious diseases are attributed to bacteria, fungi, parasites and viruses. As compared to other counterparts, bacteria are major cause of infectious diseases.

Bacteria belonging to the *Enterobacteriaceae* family are among most common human pathogens.[1] These organisms are Gram negative bacilli and are commensals of intestinal. [1] They are capable of causing a broad spectrum of clinical manifestations that range from cystitis to pyelonephritis, septicaemia, pneumonia, peritonitis and meningitis. [1, 2] Additionally, *Enterobacteriaceae* are one of the important cause of health-care associated infections (HAI). [1, 2]

Organisms belonging to *Enterobacteriaceae* family can rapidly develop resistance to antibiotics. Resistance to broad spectrum antimicrobial agents like 3rd and 4th cephalosporins is well documented in *Enterobacteriaceae*.[3]

In recent years, antibiotic resistance has become a serious threat to public health as bacteria are increasingly

developing resistance to antibiotics while drug manufacturing companies are not developing new antibiotics that can compact the menace of drug resistance.[4, 5]

Carbapenems are among the most effective antibiotics prescribed for treatment of life threatening infections caused by gram negative bacteria including *Enterobacteriaceae*. [2, 6] However in recent times, carbapenem resistance in *Enterobacteriaceae* is an ongoing public-health globally.[7, 8]

Klebsiella pneumoniae is one of the most common isolate from *Enterobacteriaceae*. It causes variety of infections like pneumonia, urinary tract infection (UTI), bacteremia, meningitis, sepsis and abscesses.[9] It is one of most common causes of HAI. In recent years, *K. pneumoniae* has developed resistance to multiple antibiotics including extended spectrum β-lactam and carbapenem antimicrobial agents.[9, 10]

In this present review article, the overview of carbapenam resistance in *K. Pneumoniae* is presented with emphasis on historical perspectives, epidemiology, laboratory identification and prevention and control.

Methodology:

For preparation of the present review article, a search was made on "Search engines" like Pubmed and Google Scholar. Literature search were made by using MeSH terms like 'carbapenem', 'carbapenemases', 'carbapenem resistance', 'carbapenem resistant *Enterobacteriaceae*' and 'carbapenem resistant *Klebsiella pneumoniae*'. Relevant 'Original Research' and 'Review' were retrieved and used for preparation of the manuscript.

Basics of carbapenems, carbapenemases and carbapenem resistance.

β-lactam group is collection of antibiotics that share a common β lactam ring.[11] These include antimicrobial agents like penicillin, cephalosporins, monobactams and carbapenems. These antibiotics bind and inactivate the penicillin-binding proteins (PBPs), which are responsible for cell wall formation in bacteria.[11] This group of antibiotics represent the most common therapeutic agent for bacterial infections.

The empirical and injudicious use and persistent exposure of bacterial pathogens to a variety of β-lactam antibiotics have resulted in emergence of extended spectrum β-lactam (ESBL) producing bacterial strains.[12] In recent years, a significant rise in isolation of ESBL producing *Enterobacteriaceae* is reported worldwide.[13, 14]

Among various members of *Enterobacteriaceae* family *E. coli* and *K. pneumoniae* are the major ESBL producers globally.[15] The rate of isolation of ESBL producing *Enterobacteriaceae* significantly varies according to healthcare setup. [12] Various national and international have reported prevalence ESBL production in *Enterobacteriaceae* to vary from 8 to 80%.[12]

The prevalence of ESBL producing organisms highly depends on antibiotic policy of hospital, carriage rate among health care workers, adherence to infection control practices and type of disinfectant used particularly in critical care areas of the hospital.

Carbapenems are the most potent wide spectrum antibiotics used for treating life threatening infections caused by gram negative bacteria like *Enterobacteriaceae* (including ESBL producers), *Pseudomonas aeruginosa* and *Acinetobacter* spp.[16] Structurally, carbapenems resemble penicillin but has additional sulphur group at C1 position.[17]

Imipenem, meropenem, ertapenem, doripenam, panipenem and biapenem are examples of carbapenems used in clinical practice.[1, 18] The cyclic amine like meropenem, doripenam, panipenam that have pyrrolindine derivatives have broad spectrum activity. [18]

Doripenam is known to have lower MIC compared to meropenem and imipenem for *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. [19, 20] However it is less susceptible and comparatively slower in hydrolysing carbapenemase than imipenem.[21]

As compared to meropenem and imipenem, ertapenem is less sensitive against *Pseudomonas aeruginosa* whereas imipenem and doripenem has good susceptibility for *Acinetobacter baumannii* compared to meropenem.[18]

All carbapenems are recommended for treating crucial and life threatening HCAIs linked to organ transplantations, ICUs and surgeries. [1, 18] As per recent research, transpeptidase inhibition is the main enzyme target of carbapenems during the process of cell wall synthesis in bacteria. [18]

Emergence of carbapenem resistant *Enterobacteriaceae* (CRE) strains is one of the major threat to the development of modern therapeutic modalities in medicine. To define, CRE are *Enterobacteriaceae* that are resistant to one or all carbapenems and all of the third-generation cephalosporins.[22]

Production of carbapenemases is the most common mechanism for carbapenem resistance. Carbapenemases are β lactamases that have capacity to hydrolyse both carbapenem and all other β lactam antibiotics.[2] Therefore, all carbapenemases are β lactamases but not all β lactamases are carbapenemases. Additionally, carbapenemases also led to overexpression of multidrug efflux pumps by the bacterial cell membrane. [2]

On the basis of the amino acid sequences, carbapenemases are broadly classified into metallo β-lactamases (Class B) and serine β-lactamases (Classes A, C and D). [17] Metallo β-lactamases contain zinc at the active site whereas serine β-lactamases contain serine at the active site. [17, 23] Class C enzymes are mostly found in gram negative pathogenic bacilli like *Acinetobacter* spp., *Aeromonas* spp. and *Enterobacter* spp. [17] These Class C enzymes are chromosomally encoded. The combination of β-lactamases like Class A and Class C confer resistance to most antibiotic classes like cephamycins, penicillins and cephalosporins.[24] They are not inhibited by inhibitors such as clavulanic acid. [17]

As per Ambler classification, β-lactamases are categorized into four molecular classes.[11] Among them, carbapenemase activity is shown by the class A enzymes KPC, GES/IBC, IMI/NMC-A, SFC-1, the Class B MBLs IMP, VIM, NDM, SPM, GIM, SIM, AIM, DIM, FIM, POM and several class D (OXA-type) enzymes.[11] Class C enzymes are not considered carbapenemases. However, this class of β-lactamases possess a low potential of carbapenem hydrolysis. [25] The over-production of Class C enzymes may contribute to carbapenem resistance in association with decreased outer-membrane permeability and/or efflux pump over-expression. [25]

In *K. pneumonia*e, carbapenemases (KPC) in association with other members like SME, IMI, NMC, GES constitute the Class A. The KPC were first described in USA. [3] In *Pseudomonas aeruginosa,* the class B enzymes constitute of active on imipenem (IMP), Verona integrin encoded MBL (VIM), SPM, GIM, New Delhi β-lactamases (NDM) and SIM families. [17] In addition to *K. pneumoniae*, Class A carbapenemase is also found in gram negative bacteria like *E. coli*, *K. oxytoca*, *Enterobacter* spp., *Serratia* spp., *Salmonella* spp., *Citrobacter freundii* and *Pseudomonas aeurignosa*.[17] Globally, among various classes of carbapenemases, production of Class A is the most common mechanism involved in resistance.

Two important mechanisms are described for carbapenem resistance in *Enterobacteriaceae*. [1] First mechanism includes acquisition of carbapenemase genes encoding enzymes that degrades carbapenems.[1] SME-1 was the first carbapenemases to be identified in *Enterobacteriaceae.* It was identified in *Serratia marcesnes* in London (1982). Later on, IMI-1 was identified in USA (1984). [17]

Second mechanism of carbapenem resistance in *Enterobacteriaceae* involves decrease uptake of antibiotics due to quantitative and/or qualitative deficiency of porin expression in combination with increased expression of β-lactamases possessing weak affinity to carbapenems.[1] The overexpression of efflux pumps expel carbapenem and finally results in carbapenem resistance. This mechanism may also confer multidrug resistance in an isolate as resistance to antibiotics like quinolones, penicillin, cephalosporins and aminoglycosides have common substrates for efflux pump. Combinations of these mechanisms result in high level of carbapenem resistance in bacterial species like *K. pneumoniae*, *P. aeruginosa*, and *A. baumannii*.[21]

Carbapenemases production is very serious and important problem in the so-called 'ESKAPEE' (*E. faecium*, *S. aureus*, *Klebsiella*, *Acinetobacter*, *Pseudomonas*, *Enterobacter*, and *E. coli*) pathogens.[26] 'ESKAPEE' pathogens collectively are major cause of HAI. [26] The Centers for Disease Control and Prevention (CDC) have underscored the importance of protocols emphasizing prevention and control of CRE transmission in health care facility.[27]

Various risk factors including ICU stay, prolonged hospitalization, pulmonary disease, hepatic disorders, immunocompromised status, antibiotic therapy, history of carbapenem usage, β-lactamas/ β-lactamases inhibitors, long term antibiotic therapy and indwelling medical devices are identified for carbapenem resistance in *Enterobacteriaceae*.[3] Rectal colonization with CRE is an important risk factor for development of CRE infections. [26]The CDC recommends perirectal screening along with isolation of patients either colonized or infected with CRE.

Factors like, extreme and injudicious antibiotic prescription, over the counter sale of antibiotics, noncompliance with infection control measures, and the use of sub-therapeutic antibiotic for the promotion of animal growth in the animal husbandries are also responsible for emergence and wide spread of CRE.[3]

Carbapenem resistance in *Klebsiella pneumoniae*.

(i) Historical perspectives.

Thienamycin was the first "carbapenem" to be discovered. It eventually served as the parent or model compound for development all carbapenems.[28] The notable discovery of Thienamycin was first reported in 1976 at the 16th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC). [28]

Thienamycin displayed good anti-microbiological activity against wide range of gram negative bacteria, including *Pseudomonas aeruginosa*.[29] It was also active against anaerobes (*Bacteroides fragilis*) and gram-positive cocci like methicillin susceptible *Staphylococcus aureus* (MSSA) and *Streptococci*. [29]

At that time, thienamycin derived compounds were of great clinical use because of emergence of cephalosporin resistant gram negative and gram positive pathogens. [28] However the stability of thienamycin was lost in aqueous solution. [28] This compound hydrolysed even at slight pH of 8.0. Thienamycin was noted to be highly reactive with nucleophiles and even with self-primary amine. [28]

In 1985, imipenem (originally known as MK0787) was the first carbapenem available for the treating complex bacterial infections. [28] It was more-stable compared to derivatives of thienamycin. [28] Imipenem was found to be less sensitive to hydrolysis in alkaline solution. Later on several carbapenems with broad spectrum of activity and more stability like meropenem, biapenem, ertapenem, and doripenem were developed. [28]

Carbapenem resistance may be innate or acquired or both. However in *K. pneumoniae* is intrinsically susceptible to carbapenems.[22] It usually acquire resistance due to carbapenem-hydrolysing β-lactamases. [17]

KPC producing *K. pneumoniae* strains were first isolated in North Carolina in 2001. In France, it was reported in 2005.[30]Later, in 2007 KPC producing *K. pneumoniae* was described in Greece and many other European countries including Belgium, Denmark, England, Italy and Scotland.[28] In Greece, these infections were related to the transfer of patients from one hospital to another. CRE was first reported in Korea in 2010. [28]

Giani *et al* reported KPC producing *K. pneumoniae* strain for first in Italy in 2009.[31] In their study the patient received this strain from a healthcare provider who came from Israel. However, a search through available literature, revealed no documented evidence of origin of infection due to KPC producing *K. pneumoniae* in Asia. [31]

(ii) Epidemiology of carbapenem resistance in *K. pneumoniae*.

Since first report of isolation of KPC-producing *K. pneumoniae* in 2001, various outbreaks and nosocomial transmission has been reported from USA. Bratu *et al* (2005) from New York reported rapid spread of carbapenem resistant *K. pneumoniae* as a new threat to antibiotic armamentarium. In their study, 1.4% of *K. pneumoniae* isolates were reported to have the bla_{kpc} gene.[32]

Leavitt *et al* (2007) and Navon-Venezia *et al* (2009) from Israel reported increased isolation of KPC-producing *K. pneumoniae*.[33, 34] In the study of Navon-Venezia *et al* (2009), pulsed-field gel electrophoresis (PFGE) analysis of KPC-producing *K. pneumoniae* from 13 different health care centers showed a clonal relationship between many isolates.[34] Some of these isolates were found to be genetically related to strains reported from outbreaks in the USA. [34]

In an annual update (year 2006-2007) of the National Healthcare Safety Network (NHSN) carbapenem resistance was reported in 10.8% of *K. pneumoniae* isolated from medical-device associated infections.[28]

Various Indian studies have reported significant prevalence of CRE. Datta *et al* (2012) from North India reported prevalence rate of 7.87%.[35] As compared to Datta *et al* (2012), Gupta *et al* (2006) and Wattal *et al* (2010) reported high prevalence rate of CRE.[35, 36, 37] In the study conducted at a tertiary care hospital of Mumbai, Maharashtra by Nair *et al* (2013) the rate of CRE was 12.26%. In addition to various wards and the ICU, a significant percentage of CRE was also noted in OPD patients.[22] The study of Nair *et al* (2013) emphasized the need for initiation of control measures against CRE at both hospital and community level. [22]

Parimala *et al* (2017) from Karnataka reported carbapenem resistance in 42.8% of *K. pneumoniae* isolated from various clinical specimens. [2]

(iii) Laboratory methods for identification of carbapenem resistant *K. pneumoniae.*

Rapid and precise identification of CRE isolated from clinical specimen is very important to initiate control measures for CRE transmission in any health-care setup. In a clinical microbiological services various methods have been used for identification of *K. pneumoniae*.

The CDC in 2009 outlined a laboratory screening protocol for identification of CRE. This method is based on the modified Hodge test and till date is validated only for *K. pneumoniae* and *E. coli*.[38]

This CDC approved protocol is based on the method suggested by Landman *et al* (2005) originally for detection of carbapenem-resistant *K. pneumoniae* in stool surveillance cultures.[39] In this method of identification of CRE, the MacConkey's agar with a 10µg meropenem disk is used. *E. coli* ATCC 25922 susceptible to carbapenems is used as control strain.

The MHT is also known as cloverleaf test. MHT is widely used as a phenotypic method for detection of carbapenemase production. This method is based on the inactivation of carbapenem (meropenem or ertapenem) by whole cells of carbapenemase-producing isolate. MHT is associated with following demerits.[40]

(i) It cannot differentiate between the types of carbapenemase involved in carbapenem resistance.[40] (ii) With this method, false-positive results have been observed with ESBL (CTX-M-type) producing strains.[41] (iii) False-negative results may occur, mainly in MBL producing strains having weak carbapenemase activity. [41] (iv)This method is unreliable for detection of New Delhi metallo-β-lactamase (NDM-1) producing *K. pneumoniae*.[42] To overcome this problem, the use of MacConkey's agar instead of Mueller-Hinton agar for susceptibility testing has been suggested. [40] With the use of MacConkey's agar the performance of testing can be improved due to increased release of periplasmic enzymes by the action of the bile salts (one of the constituent of the MacConkey's agar). [40]

By this method, up to four isolates can be tested at a time on the same plate with single merpenem disc. The isolate is considered as a carbapenemase producer when a clover-leaf type indentation at the intersection of the isolate tested and the control strain (*E. coli* ATCC 25922) within the inhibition zone is seen after overnight incubation. [40]

In the study Anderson *et al* (2007), MHT was found to be 100% sensitive and specific for detection of KPC producing *K. pneumoniae*.[43] Mathers *et al* (2014) conducted prospective evaluation two phenotypic assay (MHT and indirect carbapenemase test (ICT)) for detection of KPC producing *K. pneumoniae*.[38] In their study, ICT was found to be superior to MHT for detection of KPC producing *K. pneumoniae* isolated from clinical specimens. They also reported, ICT to be more specific and an alternative protocol for determination of CRE. [38]

Ahmed *et al* (2017) carried out point prevalence survey for screening of CRE in ICU patients. By CDC protocol, carbapenem resistance was observed in 87.5% of *Klebsiella* spp.[26] In the same study, the authors compared susceptibility results of merpenem and ertapenem for detection of CRE. [26] Meropenem detected all CRE whereas ertapenem could identify only 84.2% of CRE. Their study concluded that, meropenem is more sensitive than ertapenem for screening of carbapenem resistance developed by different mechanisms. [26]

The susceptibility of KPC producing isolates to boronic acid and its derivatives is an example of another phenotypic method for detection of CRE. [40]Boronic acid and its derivatives like phenylboronic and 3-aminophenylboronic acid are structurally similar to β lactams including carbapenems. [40] The concept of using boronic acid and its derivatives for determining KPC producing isolates was first put forward by Pasteran *et al* (2008).[44]

Phenotypic method involving use of boronic acid and its derivatives is highly accurate and simple. [40] In this method, the diameter of the inhibition zone formed around a β-lactamic disc containing boronic acid is compared with that around the corresponding β-lactamic disc without boronic acid. [40] The zone of inhibition of size ≥ 5mm around the disc containing boronic acid than in that without boronic acid is indicative of carbapenemase activity. Giakkoupi *et al* (2009) reported boronate-based assays to be ineffective in detecting KPC producing *K. pneumoniae* isolates in the case of co-production of VIM β –lactamase.[45]

CHROMagar-KPC (CHROMagar; BBL) and Brilliance CRE agar (Thermo Fisher Scientific) are examples of commercially available chromogenic media for identification of KPC producing *K. pneumoniae*. Chromogenic media were originally formulated for screening CRE in ICU patients.[40]

Arnold *et al* (2011) reported sensitivity of 100% and specificity of 98.4% compared to polymerase chain reaction (PCR) for detection of KPC producing *K. pneumoniae.* Chromogenic media are often costly but simple for use and interpretation.[46]

KPC producing *K. pneumoniae* produces blue colored colonies on chromogenic CRE agar. Brocco *et al* (2013) reported chromogenic CRE agar as 100% sensitive for detection of KPC producing *K. pneumoniae*.[47]

In the study of Ahmed *et al* (2017) chromogenic CRE agar was reported to have a perfect agreement for detection of all mechanisms of carbapenem resistance.[26] Chromogenic CRE agar detected carbapenem resistance in isolates with 24 hours of inoculation. [26]

CRE can be also detected by reference broth dilution method (BMD). Epidemiological cutoff (ECOFF) values are used to decide non-susceptibility to carbapenem. As per The European Committee on antimicrobial Susceptibility Testing (EUCAST), the minimum inhibition concentration (MIC) ≥1 µg/ml for imipenem and ≥0.5 µg/ml for meropenem and ertapenem is ECOFF for *K. pneumoniae* and *E. coli*.[40]

As the Clinical and Laboratory Standards Institute (CLSI) has not defined ECOFF, interpretation is done on the basis of clinical breakpoints (CBPs). The isolate that is either intermediate (I) or resistant (R) to at least one carbapenem as well as resistant to a third generation cephalosporin (cefotaxime, ceftazidime, ceftriaxone, cefoperazone, or ceftizoxime) should be tested further.[40]

Various genotypic methods based on *in vitro* gene amplification are also available for detection of CRE. These include genotypic techniques like polymerase chain reaction (PCR).[40] Genotypic methods are rapid compared to phenotypic methods. Many researchers have used Multiplex and real-time (RT) PCR in addition to "in-house" PCR-based techniques for the detection of carbapenemase genes *in K. pneumoniae*.[48] Melting curve step in the RT- PCR technique can be allows the accurate identification of carbapenemase gene variants.[40]

Multitude of commercial PCR, hybridization and microarray based kits for detection are available for detection of CRE. These include Hyplex MBL ID, Hyplex CarbOxa ID kits (BAG Health Care, Lich, Germany) , The Check KPC ESBL microarray and its expanded version and Check-MDR CT102 (Check-Points Health BV, Wageningen, Netherlands). [40]

Microarray technology is recent addition to the list of genotypic methods for rapid and accurate detection of various carbapenem resistance determinants. This genotypic method is used for accurate detection of multiple bla genes within a single reaction tube. [40]

Matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS) is the latest technology that has revolutionized the detection and identification of pathogens. [40, 48] MALDI-TOF MS can be also successfully utilized for detection of CRE.

Detection of carbapenemase activity by spectrophotometric method is considered the reference method for the verification of carbapenemase activity. [40, 49] In this method, carbapenemase activity in an isolate is detected spectrophotometrically by using crude or partially purified enzyme extracts and a carbapenem drug mostly imipenem. As this method require a high technical expertise and is labor extensive, its utility is mostly restricted to reference laboratories. [40, 49]

(iv) Prevention and control of carbapenem resistant *K. pneumoniae*.

Infections due to carbapenem resistant *K. pneumoniae* are common in hospitals were where infection control practices are inadequate. Both the CDC and European Society of Clinical Microbiology and Infectious Diseases (ESCMID) have published guidelines for interventions to prevent and control CRE transmission in health care

facilities.[50] These guidelines are similar to those recommended for other multi drug resistant organisms (MDRO).[51] It includes early detection, isolation or cohorting of patients colonized or infected with CRE along with enhanced compliance with infection prevention and control measures.

Each and every hospital staff should be made aware of the concept of "Clean Care is Safer Care". Environmental hygiene and equipment cleaning along with daily chlorhexidine baths to cleanse patients colonized or infected play in important role in prevention and control of transmission of carbapenem resistant *K. pneumoniae.* Judicious and restricted use of antibiotics also prevents selection of MDRO exerted by a specific agent.

References

1. Nordmann P, Dortet L, Poirel L. Carbapenem resistance in Enterobacteriaceae: here is the storm! *Trends Mol Med* 2012; 18:263-272.
2. Parimala T. Screening of carbapenem resistant Enterobacteriaceae among nosocomial isolates: A study from South India. *Int J Curr Microbiol App Sci* 2017; 6: 460-465.
3. Gupta N, Limbago B, Patel J, Kallen A. Carbapenem-resistant Enterobacteriaceae: Epidemiology and prevention. *Clin Infect Dis* 2011; 53:60-67.
4. Carlet J, Jarlier V, Harbarth S, Voss A, Goossens H, Pittet D. Participants of the 3rd World Healthcare-Associated Infections Forum. Ready for a world without antibiotics? The Pensières Antibiotic Resistance Call to Action. *Antimicrob Resist Infect Control* 2012; 1:11.
5. World Health Organization. Antimicrobial Resistance: Global Report on Surveillance; WHO Press: Geneva, Switzerland, 2014.
6. Gupta E, Mohanty S, Sood S, Dhawan B, Das B, Kapil A. Emergence resistance to carbapenems in a tertiary care hospital in north India. *Indian J Med Res* 2006; 124: 95-98.
7. Kang J, Yi J, Ko M, Lee S, Lee J, Kim K. Prevalence and risk factors of Carbapenem-resistant Enterobacteriaceae Acquisition in an Emergency Intensive Care Unit in a Tertiary Hospital in Korea: a Case-Control Study. *J Korean Med Sci* 2019; 34:e140.
8. Zhang Y, Wang Q, Yin Y, Chen H, Jin L, Gu B *et al*. Epidemiology of carbapenem-resistant Enterobacteriaceae infections: report from the China CRE network. *Antimicrob Agents Chemother* 2018; 62:e01882.
9. Sailaja B, Singh A. *Klebsiella pneumoniae*: Prevalence of ESBL Producing Clinical Isolates and their Antimicrobial Susceptibility Pattern in a Hospital Setting. *Int J Curr Microbiol App Sci* 2019; 8: 2794-2799.
10. van Duin D, Doi Y. The global epidemiology of carbapenemase-producing Enterobacteriaceae. *Virulence* 2017; 8: 460-469.
11. Meletis G. Carbapenem resistance: Overview of the problem and future perspectives. *Ther Adv Infect Dis* 2016; 3:15-21.
12. Sarojamma V, Ramakrishna V. Prevalence of ESBL-producing Klebsiella pneumoniae isolates in tertiary care hospital. *ISRN Microbiol* 2011; vol 2011, Article ID 318348: 5 pages.

13. Livermore D. Bacterial resistance: origins, epidemiology, and impact. *Clin Infect Dis* 2003; 36:S11-S23.
14. Paterson D, Bonomo R. Extended-spectrum βlactamases: clinical update. *Clin Microbiol Rev* 2005; 18: 657-686.
15. Jacoby G, Munoz-Price L. The new β-lactamases. *N Engl J Med* 2005; 352: 380-391.
16. Cornaglia G, Rossolini G. The emerging threat of acquired carbapenemases in gram-negative bacteria. Clin Microbiol Infect Dis 2010; 16:99-101.
17. Swathi C, Chikala R, Ratnakar K, Sritharan V. A structural, epidemiological and genetic overview of *Klebsiella pneumonia* carbapenemases (KPCs). *Indian J Med Res* 2016; 144:21-31.
18. Codjoe F, Donkor E. Carbapenem resistance: A review. *Med Sci* 2018; 6:1.
19. Mandell L. Doripenem: A new carbapenem in the treatment of nosocomial infection. *Clin Infect Dis* 2009; 49: S1-S3.
20. Bassetti M, Ginocchio F, Mikulska M. New treatment options against Gram-negative organisms. *Crit Care* 2011; 15:215.
21. Queenan A, Shang W, Flamm R, Bush K. Hydrolysis and inhibition profiles of β-lactamases from molecular classes A to D with doripenem, imipenem, and meropenem. *Antimicrob Agents Chemother* 2010; 54:565-569.
22. Nair P, Vaz M. Prevalence of carbapenem resistant *Enterobacteriaceae* from a tertiary care hospital in Mumbai, India. *J Microbiol Infect Dis* 2013; 3:207-210.
23. Queenan A, Bush K. Carbapenemases: The versatile beta-lactamases. *Clin microbial Rev* 2007; 20:440-458.
24. Jung H, Sang H. Carbapenem resistance in Gram negative pathogens: emerging non-metallo-carbapenemases. *Res J Microbiol* 2006; 1:1-22.
25. Quale J, Bratu S, Gupta J, Landman D. Interplay of efflux system, ampC, and oprD expression in carbapenem resistance of *Pseudomonas aeruginosa* clinical isolates. *Antimicrob Agents Chemother* 2006; 50: 1633-1641.
26. Ahmed O. Point prevalence survey for screening of carbapeneme resistant Enterobacteriaceae in ICU patients using CDC protocol and chromogenic agar. Int J Curr Microbiol App Sci 2017; 6: 1884-1893.
27. Centers for Disease Control and Prevention. Facility guidance for control of carbapenem-resistant Enterobacteriaceae (CRE)-November 2015 update CRE toolkit. https://www.cdc.gov/hai/organisms/cre/cretoolkit. Updated 2015
28. Papp-Wallace K, Endimiani A, Taracila M, Bonomo R. Carbapenems: Past, Present, and Future. Antimicrob Agents Chemother 2011; 55: 4943-4960.
29. Fainstein V, LeBlanc B, Weaver S, Bodey G. A comparative in vitro study of thienamycin. Infect 1982; 10:50–52.
30. Ben-David D, Maor Y, Keller N, Regev-Yochay G, Tal I, Shachar D *et al*. Potential role of active surveillance in the control of a hospital-wide outbreak of carbapenem-resistant *Klebsiella pneumoniae* infection. *Infect Control Hosp Epidemiol* 2010; 31:620-626.

31. Giani T, D'Andrea M, Pecile P, Borgianni L, Nicoletti P, Tonelli F *et al*. Emergence in Italy of Klebsiella pneumoniae sequence type 258 producing KPC-3 carbapenemase. *J Clin Microbiol* 2009; 47:3793–3794.
32. Bratu S, Mooty M, Nichani S, Landman D, Gullans C, Pettinato B *et al*. Emergence of KPC-possessing Klebsiella pneumoniae in Brooklyn, NewYork: epidemiology and recommendations for detection. *Antimicrob Agents Chemother* 2005; 49:3018–3020.
33. Leavitt A, Navon-Venezia S, Chmelnitsky I, Schwaber MJ, Carmeli Y. Emergence of KPC-2 and KPC-3 in carbapenem-resistant Klebsiella pneumoniae strains in an Israeli hospital. *Antimicrob Agents Chemother* 2007; 51:3026–3029.
34. Molecular epidemiology of KPC-producing Klebsiella pneumoniae isolates in the United States: clonal expansion of multilocus sequence type 258. *Antimicrob Agents Chemother* 2009; 53: 3365-3370.
35. Datta P, Gupta V, Garg S, Chander J. Phenotypic method for differentiation of carbapenemases in Enterobacteriaceae: Study from north India. *Indian J Pathol Microbiol* 2012; 55: 357-360.
36. Gupta E, Mohanty S, Sood S, Dhawan B, Das B, Kapil A. Emergence resistance to carbapenems in a tertiary care hospital in north India. *Indian J Med Res* 2006 124: 95-98.
37. Wattal C, Goel N, Oberoi J, Raveendran R, Datta S, Prasad, K. Surveillance of multidrug resistant organisms in a tertiary care hospital in Delhi, India. *J. Assoc. Physicians India* 2010; 58:32-36.
38. Mathers A, Poulter M, Dirks D, Carroll J, Sifri C, Hazen K. Clinical microbiology costs for methods of active surveillance for *Klebsiella pneumoniae* carbapenemase-producing Enterbacteriaceae. *Infect Control Hosp Epidemiol* 2014; 35:350-355.
39. Landman D, Salvani JK, Bratu S, Quale J. Evaluation of techniques for detection of carbapenem-resistant *Klebsiella pneumoniae* in stool surveillance cultures. *J. Clin. Microbiol* 2005; 43:563-5641.
40. Tzouvelekis L, Markogiannakis A, Psichogiou M, Tassios P, Daikosc G. Carbapenemases in *Klebsiella pneumoniae* and Other *Enterobacteriaceae*: an Evolving Crisis of Global Dimensions. *Clin Microbiol Rev* 2012; 25:682-707.
41. Pasteran F, Mendez T, Guerriero L, Rapoport M, Corso A. Sensitive screening tests for suspected class A carbapenemase production in species of Enterobacteriaceae. *J. Clin. Microbiol* 2009. 47:1631–1639.
42. Centers for Disease Control and Prevention (CDC). Detection of Enterobacteriaceae isolates carrying metallo-β-lactamase-United States, 2010. *MMWR Morb. Mortal. Wkly. Rep*. 2010; 59:750.
43. Anderson K, Lonsway D, Rasheed J, Biddle J, Jensen B, McDougal K *et al*. Evaluation of methods to identify the *Klebsiella pneumoniae* carbapenemase in *Enterobacteriaceae*. *J. Clin. Microbiol* 2007; 45:2723-2725.
44. Pasteran F, Otaegui L, Guerriero L, Radice G, Maggiora R, Rapoport M *et al*. *Klebsiella pneumoniae* carbapenemase-2, Buenos Aires, Argentina. *Emerg. Infect. Dis* 2008 14:1178-1180.
45. Giakkoupi P, Pappa O, Polemis M, Vatopoulos A, Miriagou V, Zioga A *et al*. Emerging *Klebsiella pneumoniae* isolates coproducing KPC-2 and VIM-1 carbapenemases. *Antimicrob. Agents Chemother* 2009; 53:4048-4050.
46. Arnold R, Thom K, Sharma S, Phillips M, Johnson J, Morgan D. Emergence of *Klebsiella pneumoniae* carbapenemase (KPC)-producing bacteria. *Southern Med J* 2011; 104:40-45.

47. Brocco F, Vitagliano D, Demetrio F. Evaluation of a new method for detecting KPC-producing bacteria based on the combination of a screening medium and a gradient diffusion system. ECCMID 2013; P690.
48. Bisiklis A, Papageorgiou F, Frantzidou F, Alexiou-Daniel S. Specific detection of blaVIM and blaIMP metallo-β-lactamase genes in a single real-time PCR. *Clin. Microbiol. Infect* 2007; 13:1201-1203.
49. Burckhardt I, Zimmermann S. Using matrix-assisted laser desorption ionization–time of flight mass spectrometry to detect carbapenem resistance within 1 to 2.5 hours. *J Clin Microbiol* 2011; 49:3321-3324.
50. Centers for Disease Control and Prevention (CDC). Guidance for control of infections with carbapenem-resistant or carbapenemase producing Enterobacteriaceae in acute care facilities. MMWR *Morb. Mortal. Wkly. Rep.* 2009; 58:256–260.
51. Dr. Mane Priyanka M. , Dr. Patil Satish R. , Dr. Mane Makarand B , Dr. Karande Geeta S , Evaluation of risk factors and detection of toxigenic *Clostridium Difficile* infection in antibiotic associated Diarrhoea , Pravara Med Rev 2019; 11(3) September – November 2019, 44-52

Date of Submission: 10 March 2020 Date of Peer Review: 06 April 2020
Date of Acceptance: 22 May 2020 Date of Publishing: 02 June 2020
Author Declaration: Source of support: Nil, Conflict of interest: Nil
Ethics Committee Approval obtained for this study? YES
Was informed consent obtained from the subjects involved in the study? YES
For any images presented appropriate consent has been obtained from the subjects: NA
Plagiarism Checked: Urkund Software

DOI: 10.36848/IJBAMR/2020/12225.51565