

Original Research Article

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Screening of Carbapenem Resistant Enterobacteriaceae among Nosocomial Isolates: A Study from South India

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ABSTRACT

Enterobacteriaceae are inhabitants of the intestinal flora. Emergence of Carbapenem resistant Enterobacteriaceae (CRE) is a serious threat to public health. They rapidly acquire and transfer drug resistance genes through plasmids and transposons and spread to community via faeco-oral route. Hence the present study was done to know the magnitude of Carbapenem resistant Enterobacteriaceae in our hospital. This study was conducted in the department of Microbiology, SIMS and RH, Tumkuru during a period from May 2016 to October 2016. A total of 106 Enterobacteriaceae isolates were obtained by various clinical samples and from different department of the hospital. These isolates were screened for carbapenem resistance by disk diffusion method according to CLSI guidelines. Prevalence of CRE in our hospital is 44. 33%. Meropenem resistance (31. 13%) was higher than Imipenem resistance among CRE isolates. In this study, CRE is more common in male (50. 90%) patients. Majority of the sample is urine (55. 26%) and Maximum number of cases was from (51. 61%) OBG department. Most common species isolated was (63. 04%) *Escherichia coli*. Carbapenem resistant Enterobacteriaceae are more prevalent in our hospital. Hence use of carbapenem should be restricted to treat severe infections.

Keywords

Antibiotic resistance,
Carbapenem,
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Introduction

Enterobacteriaceae is a family of Gram negative bacilli which can cause a wide range of community acquired and nosocomial infections including infections of the urinary and gastrointestinal tracts, pneumonia, peritonitis, meningitis, sepsis and medical device associated infections (Surinder Kumar, 2012; Wang *et al.*, 2015; Harbaik *et al.*, 2014). These organisms easily acquire and transfer drug resistance genes through plasmids and transposons (Okochi *et al.*, 2015). Carbapenem are beta lactam antibiotics and are the last resort for the

treatment of severe infections caused by multidrug resistant Gram negative bacilli (Gupta *et al.*, 2006; Roy *et al.*, 2011). Reports indicate that carbapenemases producing Enterobacteriaceae (CRE) isolates seem to be increasing in number in the last few years (Nagaraj *et al.*, 2012).

Resistance to carbapenem is mostly due to the production of carbapenemases, which are beta lactamase enzymes with a capacity to hydrolyze not only the carbapenem but also all other beta lactam agents (Mate *et al.*,

2014). It can also be due to poor binding to carbapenem to penicillin binding protein present in the bacteria, the over expression of multidrug efflux pumps by the bacterial cell membrane (Nair *et al.*, 2013). Infections caused by CRE have limited treatment options and have been associated with high mortality rates ranged from 22% -72% (Wang *et al.*, 2015 and Weinstien *et al.*, 2011). Appropriate detection of CRE is vital for patient care in order to institute correct therapeutic options (Datta *et al.*, 2012). Hence present study was done to determine the magnitude of carbapenem resistant Enterobacteriaceae in our hospital.

Materials and Methods

This prospective study was carried out in the clinical bacteriology, in the Department of Microbiology at Shridevi Institute of Medical sciences and Research Hospital, Tumkuru during a period from May 2016 to October 2016. A total of 106 clinical isolates belong to Family Enterobacteriaceae from various clinical samples were included. The distribution of the sources of the isolates were pus, urine, blood, stool, sputum and ET secretions from different wards of hospital, outpatient departments and ICUs.

All the isolates were identified as per the standard microbiological procedure and stocked in 0.2% semisolid agar until analyzed. These isolates were screened for their ability to produce carbapenamases on Muller Hinton Agar plates by Kirby – Baurer disk diffusion method as per CLSI guidelines using Imipenem (10ug) and Meropenem (10ug) disk. Both antibiotic discs were procured from Hi-media, Mumbai.

The diameter of the zones of inhibition of growth was recorded and sensitive, intermediate and resistant based on CLSI guidelines. Organisms with intermediate

levels of resistance to the antibiotics were included in the resistant organisms for final analysis. Data were analysed with Microsoft excel.

Results and Discussion

A total of 106 Gram negative bacilli belong to Enterobacteriaceae family, were isolated during the study period. Among 106 clinical isolates, 47 CRE isolates showed resistance to carbapenem by the disk diffusion method. Out of 47 (44.33%) CRE isolates, 33 (31.13%) were resistant to Meropenem and 14 (13.20%) were resistance to Imipenem (Table 1). CRE isolates more commonly seen in 28 (50.90%) male patients than 19 (37.25%) female patients (Table 2). CRE organisms most frequently isolated on microbiological cultures of 21 (55.26%) urine samples followed by 18 (54.54%) pus samples (Fig 1), majority of the CRE isolates were from 16 (51.61) OBG department next was 14 (46.66%) medicine department. (Fig 2) The different strains of CRE isolates were *Escherichia coli*, *Klebsiella spp*, *Enterobacter spp*, *Citrobacter spp*, *Proteus spp*, *Providencia spp* and *Shigella*. Maximum number of CRE isolates in this study was 21 (55.26%) *Escherichia coli* (Table 3).

Microbial drug resistance is a growing problem of global magnitude (Praveen *et al.*, 2010). Emergence and dissemination of Carbapenem resistance among Enterobacteriaceae poses a considerable threat to public health and thought to be due to high antibiotic use and misuse without proper diagnosis of infection or self medication by patients (Okoche *et al.*, 2015; Birgy, 2012). Additionally, other factors may include co selection by other antibiotics (e. g. cephalosporins), spread of those bacteria in community, uncontrolled spread in the hospital environment, owing to undiagnosed carriers and other, still unknown, factors

(Harbaik *et al.*, 2014). The detection of Carbapenems producers in clinical infections is based first on susceptibility testing results obtained by disk diffusion / by automated systems (Nordaman *et al.*, 2011). In the present study 33 (31. 13%) were more

resistant to Meropenem than to 14 (13. 20%) Imipenem. Similarly, study conducted by Temple *et al.*, Meropenem resistance (57%) was higher than Imipenem resistance (49. 5%). This may be due to increased used of meropenem in this area (Temple *et al.*, 2015).

Table.1 Resistance to carbapenem among Gram negative bacilli belong to Family Enterobacteriaceae

Antibiotic	Carbapenem resistant	percentage	Carbapenem sensitive	percentage	Total	percentage
Imipenem	14	13.20	92	86.79	106	100
Meropenem	33	31.13	73	68.86	106	100
Total	47	44.33	59	55.66	106	100

Table.2 Gender wise distribution of CRE isolates

Gender	Carbapenem resistant	percentage	Carbapenem sensitive	percentage	Total	percentage
Male	28	50.90	27	49.09	55	51.88
Female	19	37.25	32	62.74	51	48.11
Total	47	44.33	59	55.66	106	100

Table.3 Distribution of species of CRE isolates

Species	Carbapenem resistant	percentage	Carbapenem sensitive	percentage	Total	percentage
<i>Escherichia coli</i>	29	63.04	17	36.95	46	43.39
<i>Klebsiella spp</i>	12	42.85	16	57.14	28	26.41
<i>Enterobacter spp</i>	03	27.27	08	72.72	11	10.37
<i>Citrobacter spp</i>	01	10.00	09	90.00	10	9.4
<i>Proteus spp</i>	01	16.66	05	83.33	06	05.66
<i>Providencia spp</i>	01	25.00	03	75.00	04	03.77
<i>Shigella spp</i>	00	00	01	100	01	00.94
Total	47	44.33	59	55.66	106	100

Fig.1 Sample distribution of CRE isolates

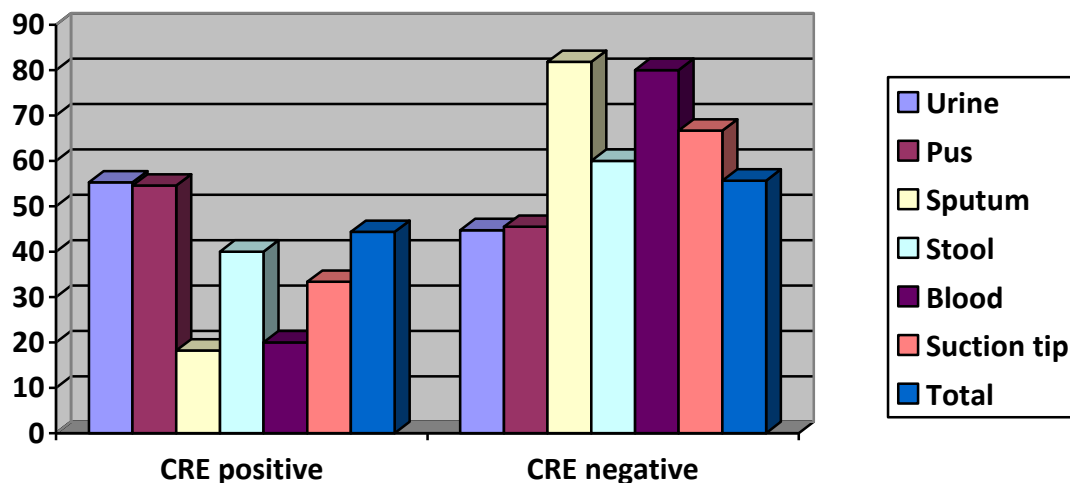
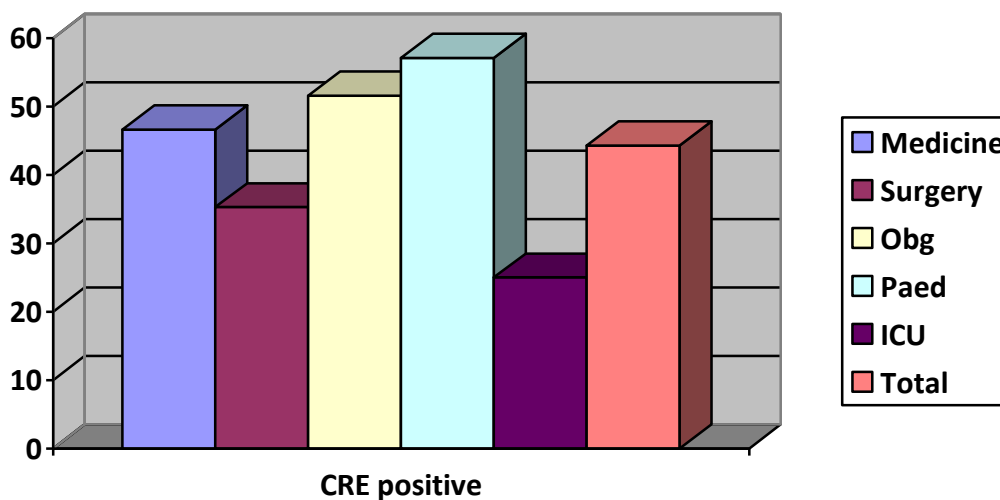


Fig.2 Distribution of CRE isolates in different department of hospital



Maximum number of CRE isolates was from 21 (55.26%) urine sample followed by pus sample (55.54%), sputum (18.18%) and others observed in our study. Nagaraj *et al.*, (2012) observed that carbapenem-resistant organism were isolated mainly from urine samples up to 42% (n=21), followed by wound discharge 18% and respiratory secretions 16%. The reason for this could be urinary tract infection (UTI), being the most common hospital acquired infection which accounts for almost 40% of all nosocomial infections (Mate *et al.*, 2014).

The most common species among CRE isolates was *E. coli* (63.04%) in the present study. Wattal *et al.*, (2010) reported high prevalence of resistance to carbapenems ranging from 13 to 51% in *E. coli* and *Klebsiella spp.* The actual prevalence of carbapenemase producers is still unknown because many countries that are likely to be their main reservoirs have not established any search protocol for their detection (Nordaman *et al.*, 2011). In the present study, prevalence of CRE is 44.33% which is higher compared to other studies. Gupta *et al.*, (2006) also

reported high prevalence of resistance varying from 17 to 22% to various carbapenems among Enterobacteriaceae strain. Datta *et al.*, (2012) found that the prevalence of CRE in their Institute is 7. 87. In the present study of prevalence of CRE is 44. 33% which is higher compared to other studies (Mate *et al.*, 2014 and Datta *et al.*, 2012).

Center for disease control has laid down guidelines for prevention of CRE infection (CDC guidelines, 2014). It includes 8 core measures which have been included in the prevention strategies and include laboratory detection; rigorous implementation and monitoring of infection control measure such as hand hygiene, contact precautions, isolation, setting up of ICC, proper medical waste disposal, and maintenance of environmental hygiene; education of training of health care personnel on proper use and rationale of infection control measures along with appreciation/reward of units/staff for best infection control; Minimal use of invasive devices; antibiotic stewardship; effective and rational dosing of antibiotics integrating pharmacokinetic and pharmacodynamic profiles of the antibiotic with the microbiological data (minimal inhibitory concentration); screening (rectal and perirectal) sampling of epidemiology linked contact of colonised or infected patients, active surveillance; and coordinated effects among health care faculties and encouragement of antimicrobial research and development.

In conclusion the present study indicates the prevalence of Carbapenem resistant Enterobacteriaceae was high (44. 33%) compared to other studies. Hence, strict implementation of infection control of policies is required to control the spread of CRE infection and Carbapenem should be used as reserve drug in the treatment of severe gram negative infections. Also, it is advisable

to use those agents such as colistin, tigecycline, fosfomycin as salvage therapy in treatment of MDR and PanDR resistance organism.

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