Prevelance of Extended Spectrum of B-Lactamases and Metallo Beta Lactamases Producing Bacteriological Isolates with Correlative Epidemiological Study of ESBLS and MBLS in MGM Hospital

G.V. Padmaja[1], R. Kondal Rao[2]

ABSTRACT

Introduction: Extended spectrum of Beta lactamases and metallo Beta lactamases producing resistant bacteria in nosocomial infections was increasing the morbidity and hospital expenditures. The source of infection is either nosocomial or community acquired or health care personnel. Comprehensive studies of source for hospital acquired infection will facilitate the control of hospital acquired infections.To emphasize the formation of hospital infection control committee (HICC) in tertiary care and teaching hospitals, computerizing and preserving antibiotic resistant data in nosocomial infections and documenting the guidelines for antibiotic policy.

Material and methods: Pus, urine, sputum and blood samples from outpatient and inpatients of MGM Hospitals of Dept. of Microbiology during January-2014 to April 2014 were processed by standard laboratory procedure as per CLSI. Anti microbial susceptibility testing was done by Kirby-Baure's disk diffusion method. ESBLs producing strains were detected by double disk synergy test after initial screening with 3rd generation cephalosporins. MBL Producing organisms were detected by double disk synergy test with Imipenam and EDTA disks. Different swabs collected from NICU, AMC, Female surgical ward, Casuality, ICCU, ENT ward, Burns ward and Central Sterilization Department are processed by standard laboratory procedure as per CLSI guidelines. ESBLs producing strains were detected by double disk synergy test after initial screening with 3rd generation cephalosporins. MBL Producing organisms were detected by double disk synergy test with Imipenam and EDTA disks.

Results: Out of 217 blood culture positives 107 were ESBL (86) AND MBLS (21). Out of 240 sputum samples positive (72) were resistant bacterial strains ESBLS (49) and (23) were MBLS. Out of 185 pus culture samples (47) were resistant strains and ESBLS (35) and MBLS (12). Out of 88 swabs collected from various wards in MGM Hospital 30 were positive from resistant strains like 8 (ESBLS) and 3 MBLS and Vancomycin resistant staphylococci were 11 and remaining 8 were sensitive to routine antibiotics.

Conclusion: The Study reveals that serious therapeutic and epidemiological spread of ESBLS and MBLS in Patients, Hospital environment hence the necessity of formation of Hospital Infection Control Committee (HICC) and to record the different patterns of antibiotic resistance in the tertiary hospitals and to upload the save in WHO NET will control the global emergence of MDR bacterial strains.

Keywords: Extended spectrum of betalactamases, (ESBLS), Metallobetalactamases, ((MBLS) Hospital Infection Control Committee (HICC) Hospital acquired infections (HAI).

INTRODUCTION

Extended spectrum of Beta lactamases and metallo betalactamases producing resistant bacteria in nosocomial infections were increasing the morbidity and hospital expenditures. The source of infection is either nosocomial or community acquired or health care personnel. Comprehensive studies of source for hospital acquired infection will facilitate the control of hospital acquired infections.[1,2] Resistant genes in bacteria confer an evolutionary advantage for survival and these traits are often associated with the emergence of multi drug resistant isolates.[3] These bacteria harbor extended spectrum of beta lactamases and metallo beta lactamases turn into harmful super bugs. Hence detection of ESBl's and MBLS and documenting their prevalence in patients, healthcare personnel and hospital environment will decrease the morbidity and hospital expenditures.[4] Over the last 25 years with each new class of new beta lactum antibiotics, new beta lactamases emerged which causes resistance to the particular class of drugs. These beta lactamases have been found in many different genera of Enterobacteriaceae and P. aeruginosa.[3] Currently, no standardized method for MBL detection has been proposed, and despite PCR being highly accurate and reliable, its accessibility is often limited to reference laboratories. Several nonmolecular techniques have been studied, all taking advantage of the enzyme's zinc dependence by using chelating agents, such as EDTA.[6-8] Early phenotypic detection of ESBLs and MBLs carrying organisms including those with susceptibility to 3rd and 4 th generation cephalosporins and also to carbapenams is of paramount importance as it allows rapid initiation of strict infection control practices as well as therapeutic guidance for confirmed infection.And this study will focus on characterization of ESBl's and MBLS, and the importance of detection of these enzymes and their epidemiology.[5]

Aims of study

- To isolate and identify the ESBLS and MBLS producing bacteria from different bacteriological cultures.
- To correlate these organisms isolated from different bacteriological cultures with isolates from nosocomial or community acquired on health care personnel.
- To emphasize the importance of formation of Hospital Infection Control Committee (HICC) in teaching and

[1]Assistant Professor, [2]Professor and Head, Department of Microbiology, Kakatiya Medical College Warangal-506004, India

Corresponding author: Dr. G. V. Padmaja, Assistant Professor, Department of Microbiology, Kakatiya Medical College Warangal-506004, India

How to cite this article: G.V. Padmaja, R. Kondal Rao. Prevelance of extended spectrum of B-lactamases and metallo beta lactamases producing bacteriological isolates with correlative epidemiological study of ESBLS and MBLS in MGM hospital. International Journal of Contemporary Medical Research 2016;3(6):1769-1771.

S. No.	Type of Culture Specimen	No.of Culture Specimens	No. of Positive Cultures	Sensitive to Routine Antibiotics	Resistant to Routine Antibiotics	DDST Positive for ESBLs	DDST Positive for MBLs
1.	Blood	740	217	110	107	86	21
2.	Urine	315	39	22	17	14	3
3.	Sputum	240	158	86	72	49	23
4.	Pus	185	72	25	47	35	12

Table-1: Results of showing the distribution Pattern of Positive Cultures Tests

S.No.	Resistant Strains	DDST Positive for % ESBLS	Percentage of ESBLS	DDST Positive MBLS	Percentage of MBLS.
1.	107	86	80.3%	21	19.7%
2.	17	14	82.3%	3	17.7%
3.	72	49	68.1%	23	31.9%
4.	47	35	74.5%	12	25.5%

Table-2: Results showing the Percentage of ESBLS and MBLS in Culture Positive Resistant Strains

S. No.	Name of the Ward	No. of Swabs Taken	No. of Swabs Showing Culture Sterile	No. of Swabs Showing Culture Positive
1.	NICU	8	7	1
2.	AMC	6	4	2
3.	FSW	8	8	3
4.	FMW	10	8	2
5.	Casuality	10	5	5
6.	MMW	9	7	2
7.	ICCU	10	9	1
8.	ENT	8	6	2
9.	CSD	9	5	4
10.	Burns Wards	10	2	8
Total		88	58	30

Table-3: Showing results of cultures in surveillance swabs

tertiary care hospitals.

- Computerizing and preserving the antibiotic resistant data in nosocomial infections which can be analyzed by all departments' clinicians which can act as a guide line for drafting an antibiotic policy.
- Uploading the same information in WHO NET that will control the global emergence of multidrug resistant strains.

MATERIAL AND METHODS

Total 740 blood culture samples, 315 urine culture samples, 240 Sputum culture samples and 185 pus culture samples were received in bacteriological section of department of microbiology, MGM hospital during January 2014 to April 2014 processed by standard laboratory procedure as per CLSI guidelines.[2] Anti microbial testing was done by Kirby – bauers disk diffusion method as a screening test ESBL testing was done by using Double disk synergy test (DDST) as per CLSI guidelines.[3] Ceftazidime (30 µgm) with and without Clavulanic acid (10 µgm) were used.

The isolates which are resistant to 3rd generation Cephalosporins with Clavulanic acid and imipenam (10µgm) are subjected to Double Disk Synergy Test imipenam with EDTA to detect metallo beta lactamase activity.[6] These imipenam EDTA disks were prepared by 2 methods.

The combined disk assay employs a B lactam disk usually a imipenam or ceftazidime to which an MBL inhibitor (IMBL) is added by 2 methods.[7-9]

1. Added the IMBL solution directly on imipenam disk already placed on the Agar plate (AD).
2. Or with previously prepared disk (PP).There were clear cut increased inhibition zones in Double Disk Synergy Tests (DDSTS)

Different swabs collected from NICU, AMC, Female surgical ward, Casuality, ICCU, ENT ward, Burns ward and Central Sterilization Department were processed by standard laboratory procedure as per CLSI guidelines. ESBL and MBL Producing Bacteriological isolates were subjected to Double Disk Synergy Tests (DDSTs).

STATISTICAL ANALYSIS

Descriptive statistics like mean and percentage were used to infer data. Microsoft office 2007 was used to make tables.

RESULTS:

Out of 740 Blood culture samples,217 Culture positive strains identified and 107 were resistant bacteriological strains to 3rd generation cephalosporins and identified 86 were ESBL Producing and 21 were MBL Producing strains as shown in Table-1. Out of 315 urine culture samples, 39 were culture positive and 17 were detected as resistant. Bacteriological strains with 14 ESBL producing strains 3 MBL producing strains. Out of 240 sputum sample 158 were culture positive and 72 were detected as resistant bacteriological strains with ESBLS 49 and MBLS were 23. Out of 185 pus culture samples 72 were detected as positives and 47 were resistant bacteriological s-trains with 35 ESBLS and MBLS were 12 as shown in Table-1 and Table-2. Out of 88 swabs collected from NICU, AMC, Female surgical ward, Casuality, ICCU, ENT ward, Burns ward and Central Sterilization Department, 30 among 88 swabs were culture positive 7 were positive from patients beds,6 were positive from health care personnel,4 positive from medicine trays and 13 were positive from walls and floors of Different wards, mainly from Burns wards. Out of 30 positive cultures 11 were resistant strains ESBLS (8) and MBLS (3), Vancomycin resistant Staphylococci were (11) and remaining (8) were sensitive to routine antibiotics as shown in Table-3.

Most commonly isolated organisms Klebsiella, Proteus, Pseudomonas, E.coli, Coagulase positive staphylococci, Coagulase negative staphylococci and Streptococcus pneumonia.[7,9,10]

Out of 30 positive cultures 7 were positive from patients' beds, 6 were positive from health care personnel, 4 were positive medicinal trace and 13 were positive from walls and floors of different wards mainly from burns wards. Out of 30 positive cultures 11 were resistant strains ESBLs (8) 26.67% and MBLs (3) 10% and Vancomycin resistant staphylococci were 11 (36.6%) and remaining 8 (26.67%) were sensitive to routine antibiotics as shown in Table-3.

DISCUSSION

In this study among the resistant strains isolated from blood ESBLs were around 80.3% and MBLs were 19.7%. Among resistant strains isolated from urine culture ESBLs were 82.3% and MBLs were 17.7%. In sputum cultures the resistant strains show 68.1% and 31.9% MBLs. Out resistant strains obtained from pus culture ESBLs were 74.5% and MBLs were 25.5%.[1-3] This indicates the need of intense surveillance in institution to control the spread of resistant strains in hospital acquired infections and necessity of formation of HICC (hospital infection control committee) in teaching hospitals and also in tertiary care hospitals.[4-6]

The British Society of Antimicrobial Chemotheraphy and Health Protection Agency of United Kingdom suggest testing of all isolates of gram negative bacteria with Ceftazidime (the best indicator for TEM and SHV derived ESBLs) and Cefotaxime (the best indicator for CTX-M types) or with Cefopodoxime (a good indicator for all ESBL types) as a first screening test. Girlyapur et. al. showed that Ceftazidime to have better sensitivity and specificity as a screening agent for efficient detection. PCR is not available in this institute. Ceftazidime, Cefotaxime and Cefopodoxime all drug disks were used for screening of ESBLs. Imipenam EDTA disks were used for confirmation of MBL producers.[7-10]

CONCLUSION

The Study reveals that serious spread of ESBLs and MBLs inpatients, hospital environment and health care personnel. Judicious use of antibiotics using carbapenams is essential to prevent hospital acquired infections. Hence the necessity of formation of Hospital Infection Control Committee (HICC) in all teaching hospitals and tertiary care hospitals and to record the different patterns of antibiotic resistance and ready availability of information in WHO NET will control the global emergence of MDR bacterial strains.

REFERENCES

1. Bradford PA. Extended spectrum β- lactamases in the 21st century: characterization, epidemiology, and detection of this important resistance threat.Clin Microbiol Rev 2001;14:933-51.
2. Clinical laboratory standards institute. Performance standards for antimicrobial susceptibility testing. Sixteenth international supplement. CLSI document M100 – S 16, Wayne PA: 2007.
3. Jarlier V, Nicolas MH, Fourneir G. Extended spectrum β-lactamases conferring transferable resistance to newer β-lactam agents in Enterobacteriaceae: Hospital prevalence and susceptibility patterns. Rev Infect Dis. 1998;10:867-78.
4. Varsha Gupta. An update on newer b-lactamases. Indian J Med Res. 2007;126:417-427.
5. Craven E, Chroneou A, Principles and Practice of Infectious Diseases 7th ed. London: Elsevier Churchill Livingstone 2005.
6. Franklin, C., L. Liolios, A.Y. Peleg. Phenotypic detection of carbapenem-susceptible metallo-β-lactamase-producing gram-negative bacilli in the clinical laboratory. J. Clin. Microbiol. 2006;44:3139-3144.
7. Lee, K., Y. Chong, H.B. Shin, Y. A. Kim, D. Yong, and J.H. Yum. Modified Hodge and EDTA-disk synergy tests to screen metallo-beta-lactamase-producing strains of Pseudomonas and Acinetobacter species. Clin. Microbiol. Infect. 2001;7:88-91.
8. Marchiaro, P., M.A. Mussi, V. Ballerini, F. Pasteran, A.M. Viale, A.J. Vila, and A.s. Limansky. Sensitive EDTA-based microbiological assays for detection of metallo-β-lactamases in nonfermentative gram-negative bacteria. J. Clin. Microbiol. 2005;43:5648-5652.
9. Migliavacca, R., J.D. Docquier, C. Mugnaioli, G. Amicosante, R. Daturi, K. Lee, G.M. Rossolini, and L. Pagani. Simple microdilution test for detection of metallo-β-lactamase production in Pseudomonas aeruginosa. J. Clin. Microbiol. 2002;40:4388-4390.
10. Petropoulou, D., K. Tzanetou, V.P. Syriopoulou, G. L. Daikos, G. Ganteris, and E. Malamou-Lada. Evaluation of imipenem/imipenem+EDTA disk method for detection of metallo-beta-lactamase-producing Klebsiella pneumonia isolated from blood cultures. Microb. Drug Resist. 2006;12:39-43.
11. Yong, D., K. Lee, J.H. Yum, H.B. Shin, G.M. Rossolini, and Y. Chong. Imipenem-EDTA disk method for differentiation of metallo-β-lactamase-producing clinical isolates of Pseudomonas spp. And Acinetobacter spp. J. Clin. Microbiol. 2002;40:3798-3801.

Source of Support: Nil; **Conflict of Interest:** None

Submitted: 25-04-2016; **Published online**: 30-05-2016