

Detection of ESBL (extended spectrum β lactamases) from urinary isolates of multi drug resistant enterobacteriaceae in a tertiary care hospital in Mangalore

Lumbeni Kithan^{1*}, Thomas S Kuruvilla², Tina Damodar¹

¹Post Graduate Resident, ²Associate Professor, Department of Microbiology, Father Muller Medical College, Mangalore, 575002, INDIA.

Email: lumbeni0011@gmail.com

Abstract

Introduction: The incidence of Extended Spectrum β Lactamase (ESBL) producing strains is increasing over the years. ESBL producing organisms pose problem for treatment. ESBL producers are also higher in uropathogens and baseline knowledge of ESBL organisms is mandatory for formulating control measures hence an attempt was therefore made to study ESBL production from urinary samples at a tertiary care hospital at Mangalore. **Aim:** To detect the number of ESBL producers among uropathogens in our centre and to compare two methods of ESBL detection. **Materials and Methods:** 1121 urinary samples were processed, of this 319 were Multi-drug resistant and were tested for ESBL production by Double Disc approximation and CLSI Confirmatory Test. **Results:** Out of 319 MDR samples, 153 were Enterobacteriaceae 93 were positive for ESBL i.e., 60.78%. Inpatients were more than outpatients and the two methods of ESBL detection were comparable. In this study ESBL was 60.78% among MDR enterobacteriaceae and enterobacteriaceae being the commonest organism in UTI, an attempt to contain ESBL organism may have some impact on decreasing the load of MDR organism. **Conclusion:** The burden of ESBL's among Enterobacteriaceae uropathogens continues to pose a challenge in treatment. Carbapenems were a mainstay in treating these cases and either methods of ESBL detection may be conveniently used. However attempts to minimize ESBL spread through hygiene, contact precautions, suitable antibiotic policy to include cheaper and more effective antimicrobials are the need of the hour. **Key words:** CLSI confirmatory test, double-disk diffusion, ESBL, multidrug resistance.

*Address for Correspondence:

Dr. Lumbeni Kithan, Resident, Department of Microbiology, Father Muller Medical College, Mangalore 575002, Karnataka, INDIA.

Email: lumbeni0011@gmail.com

Received Date: 10/09/2014 Accepted Date: 19/09/2014

Access this article online	
Quick Response Code:	Website: www.statperson.com
	DOI: 10 October 2014

INTRODUCTION

Unlike the developed countries, India like other developing countries has a heavy burden of MDR (Multi drug resistance) Organism, and many of the MDR are caused by ESBL (extended spectrum β lactamases) producers.^{1,2,3} In some studies in India, ESBL percentage is 62-100% among E-Coli and Klebsiella.¹ ESBL

enzymes hydrolyse β Lactam rings and hence resistant to cephalosporins, aztreonam but are inhibited by Clavulanic acid.² UTI is one of the most common infections^{4,5} for which antimicrobial agent is usually prescribed.⁶ Enterobacteriaceae are leading cause in Urinary tract infection (UTI).^{6,7,8} ESBL producers are also higher in uropathogens.⁸ Hence detection of ESBL from enterobacteriaceae was attempted. This part of India particularly coastal areas is geographically different from main land India and still very little data exists.³ Studies suggest the need for area or institution based approach for screening or intervention if needed, for this baseline knowledge of ESBL organisms is mandatory, without which control measures cannot be formulated.³ ESBL producers from community are also on the rise making it a public health concern⁸ studies suggest, that ESBL positive patient in UTI had longer hospital stay and more symptoms⁹, hence in this studies the true burden of ESBL

enterobacteriaceae in urine culture positive sample and MDR organism was attempted.

MATERIALS AND METHODS

The study was conducted at the Department of Microbiology. After ethical clearance, 1121 urinary samples from Sep 2013 to Nov 2013 that were clinically suspected for UTI were processed. Out of this 472 were reported with sensitivity. From this 319, samples that were MDR (Multi Drug Resistance) i.e. resistant to 1st, 2nd, 3rd generation cephalosporin and 153 were Enterobacteriaceae, these 153 samples were further processed for ESBL detection.

Antibiotic Susceptibility testing

The isolates were tested as per CLSI guidelines.¹⁰ The drug discs used for screening were Amoxicillin/Clavulanic acid (Amoxclv) (20/10µg), Cefazolin (30µg), Cefuroxime (30µg) and Cefotaxim (30µg), for ESBL production, after standardization to 0.5 Mc Farland.

1. Double Disc approximation

The organism was swabbed on Mueller Hilton agar. Amoxclv (20/10µg) and Cefotaxim (30µg) discs were placed 15mm apart and incubated (18-24 hrs). Organisms showing extension towards Amoxclv disc were considered as ESBL producers. [fig :1]

2. CLSI Confirmatory Test

Ceftazidime (30µg) and Ceftazidime/clavulanate (30/10µg) discs were placed and organisms that showed more than 5mm increase in zone on the Ceftazidime – clavulanate were considered ESBL producers. [fig:1]

RESULT

Out of the 1121 urine samples processed 472 were culture positive, among this 319 were MDR; 153 of MDR organism were enterobacteriaceae and were further tested for ESBL production; of which 93 Enterobacteriaceae were positive ESBL production. [Table 1]

Only 2 samples out of 93 showed negative by Double disc but positive by CLSI confirmation test.

Table 1

Age	MDR +ve	ESBL +ve
Birth -18	11	8
19-40	43	28
41-60	48	28
61-90	51	29
TOTAL	153	93

Table 2

IP	OP
MDR – 120	33
ESBL - 73	20

Table 3

Strain/Sex	Male	Female
MDR	65	88
ESBL	39	54

Among the total MDR enterobacteriaceae urine isolates, IP (In patient) sample were more than OP (outpatient) sample. [Table 2] The age group ranged from 37 days to 86 years, and maximum no of samples were from the older age (above 61 years) and least from less than 18 years [Table 1]. Females were more than males for both MDR and ESBL [Table 3].

DISCUSSION

Our findings of ESBL enterobacteriaceae is 60.78% which is similar to previous studies in this area Among the Enterobacteriaceae, the most predominant ESBL producers was E.coli followed by Klebsiella pneumonia. This was in contrast to a study by T.Menon et al where enterobacter was predominant. Carbapenems were 100% sensitive and the mainstay in treatment of these cases by T.Menon *et al.*¹⁴ (6.6 -68%) (11, 12, 13). MDR positive samples were 67.58% (319 out of 472). There was no significant difference between the two methods of ESBL detection. Double Disc diffusion as compared to CLSI confirmatory test statistically showed a sensitivity of 97.84%; hence may be concluded that both methods are equally effective for detecting ESBL. Some studies find MDR more in older age group.⁶ In this study though MDR was more in older age group; p value was 0.95 i.e. not significant MDR and ESBL were more in IP's as compared to OP's which is similar to other studies.^{4,6} In this study ESBL was 60.78% among MDR enterobacteriaceae and enterobacteriaceae being the commonest organism in UTI¹³ an attempt to contain ESBL organism may have some impact on decreasing the load of MDR organism. ESBL producers are increasing throughout the world¹⁵ and studies associate it with increase use of antibiotic.^{16,17} UTI is a common bacterial infection and antibiotics are usually prescribed prior to culture result,^{6,18} also UTI is frequently misdiagnosed¹⁸

CONCLUSION

In this study both the method of ESBL detection were comparable hence any method may be conveniently used. The ESBL load was comparable to other studies done in this area hence some policy to contain ESBL in this area if effective may be adopted here also. An effective antibiotic stewardship will go a long way to contain this menace of ESBL's among uropathogens.

REFERENCES

1. Manoharan A, Premalatha K, Chatterjee S, Mathai D. Correlation of TEM, SHV & CTX-M extended-spectrum beta lactamases among Enterobacteriaceae with their in vitro antimicrobial susceptibilities. *Indian journal of medical microbiology* 2011;29(2):161-4.
2. Taneja and Sharma . ESBLs detection in clinical microbiology: why & how ? *Indian J Med Res* 127 2008;April: 297-300.
3. Bhattacharya S. Is Screening patients for antibiotic resistant bacteria justified in the Indian context ? *Indian journal of medical microbiology* 2011;29(3):213-17.
4. Fouquet M, Morange V, Bruyere F. Five years follow-up of infections with extended-spectrum beta-lactamase producing enterobacteriaceae]. *Prog Urol* 2012;22(1):17-21.
5. Maya AS, Prabhakar K and Sarayu YL. A Study on Prevalance & Evaluation of Clinical Isolates from Community Acquired Infections using Different Media in Semiurban areas. *World J. Med. Sci* 2010;5(2):49-53.
6. Fircanis S, McKay M. Recognition and Management of Extended Spectrum Beta Lactamase Producing Organisms (ESBL). *Geriatrics for Practicing Physician* 2010;93:161-2.
7. Nahum KC, Odes LS, Riesenber K, Schlaeffer F and Borer A. Urinary Tract Infections Caused by Multi-Drug Resistant *Proteus mirabilis*: Risk Factors and Clinical Outcomes. *Springer Link* 2010;38:41-46.
8. Tankhiwale SS, Jalgaonkar SV, Ahamad S, Hassani U. Evaluation of extended spectrum beta lactamase in urinary isolates. *Indian J Med Res* 120 2004;Dec: 553-56.
9. Briongos-Figuero LS, Gomez-Traveso T, Bachiller Luque P, Dominguez-Gil Gonzalez M, Gomez-Nieto A, Palacios Martin T et al. Epidemiology, risk factors and comorbidity for urinary tract infections caused by extended-spectrum beta-lactamase (ESBL)-producing enterobacteria. *Int J Clin Pract* 2012;66(9):891-96.
10. Ferraro MJ, Swenson JM. Clinical and Laboratory Standard Institute, 2009. *Performance standards for antimicrobial susceptibility testing: nineteenth informational supplement*. CLSI document M100-S19. Vol 29(3).
11. Shiju M P, Yashavanth R, Narendra N. Detection Of Extended Spectrum Beta-Lactamase Production And Multidrug Resistance In Clinical Isolates Of *E.Coli* And *K.Pneumoniae* In Mangalore 2010; June (4): 2442 – 2445.
12. Rao SPN, Rama P S, Gurushanthappa V, Manipura R, Srinivasan. Extended-spectrum beta lactamases producing *Escherichia coli* and *Klebsiella pneumoniae* : A Multi-centric Study Across Karnataka K. 2014 ; 6 (1): 7-13.
13. Nair TB, Bhat GK, Pai V, Shantharam M . Extended spectrum β lactamase (esbl) in uropathogenic escherichia coli, prevalence and susceptibility pattern in south indian city. *IJRAP* 2011;2(6):1756-57.
14. T Menon, D Bindu, CPG Kumar, S Nalini, MA Thirunarayan. Comparison of double disc and three dimensional methods. *Indian Journal of Medical Microbiology* 2006;24 (2):117-120.
15. Babypadmini S, Appalaraju B. Extended Spectrum β Lactamases in Urinary isolates of *Escherichia Coli* and *Klebsiella Pneumoniae*-Prevalence and susceptibility Pattern in Tertiary Care Hospital. *Indian Journal of Medical Microbiology* 2004;22(3):172-74.
16. Tantry BA, Rahiman S. TO SCREEN FOR ESBL PRODUCERS IN A TERTIARY CARE HOSPITAL. Antibacterial resistance and trend of urinary tract pathogens to commonly used antibiotics in Kashmir Valley. *West Indian med. J*; 61 (7) : *Print version* ISSN 0043-3144.
17. Kumar MS, Lakshmi V, Rajagopalan R. Related articles, occurrence of extended spectrum beta-lactamases among Enterobacteriaceae spp. isolated at a tertiary care institute. *Indian J Med Microbiol* 2006; 24: 208–11.
18. Hecker MT, Fox CJ, Son AH, Cydulka RK, Siff JE, Emerman CL, Sethi AK, et al. Effect of a Stewardship Intervention on Adherence to Uncomplicated Cystitis and Pyelonephritis Guidelines in an Emergency Department Setting. *PLoS ONE* 2014 ; 9 (2): e87899.

Source of Support: None Declared
Conflict of Interest: None Declared