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Brief Communication

Incidence of extended spectrum beta lactamase producing *Escherichia coli* among patients, healthy individuals and in the environment

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Abstract

We investigated the faecal carriage of extended spectrum β-lactamases (ESBL) producing *Escherichia coli* in different groups of human subjects and in the environment. A total of 363 *E. coli* strains were isolated from stool samples of patients (n = 77), healthy subjects (n = 170) and from different environmental samples (n = 116). A total of 124 ESBL producing *E. coli* strains were isolated in this study. The frequency of ESBL producing *E. coli* was found to be highest (60.3%) among the strains isolated from patients, followed by healthy individuals (38%) and the environment (10.5%). The environment was observed to have a very low number of ESBL producing *E. coli.*

Key words: *Escherichia coli, extended spectrum* β*-lactamase, resistance, susceptibility*

Introduction

Relentless use of β-lactam antibiotics in the clinical practice has resulted in the appearance of newer β-lactamases such as extended spectrum β-lactamases (ESBLs), are typically plasmid mediated and seen mainly in *Escherichia coli and Klebsiella pneumoniae.* Among the many ESBL enzymes characterised, temoneira (TEM), sulfhydryl variable (SHV) and CTX-M are the commonly reported ones.[1] CTX-M type ESBL is emerging rapidly in many parts of the globe. The CTX-M enzyme coding plasmids spread and the strains cause outbreaks both in hospitals and in the community.[2,3] Our study was undertaken to analyse the frequency of faecal carriage of ESBL producing *E. coli* in patients and among healthy individuals and in the environment.

Materials and Methods

The study was carried out between July 2011 and September 2012. Faecal samples were collected from patients admitted in wards and high dependency units (HDUs) of our study hospital with a minimum stay of 4 days and on antibiotic treatment. Faecal samples were collected from healthy adults and infants (age 0-2) with no history of major infective illness and no antibiotic usage in the recent past (3 months). Environmental sampling such as sewage water from various sewage drains, swabs from public toilets, market places and slaughter houses and fish shops in Vellore were included. The hospital environment was studied by collecting swab samples from wards, procedure rooms and intensive care units of the study hospital. Samples from the hospital sewage were also included in the study. The faecal samples were processed and identified by standard methods.[4]

All the *E. coli* isolates were screened for ESBL production using double disk method according to Clinical Laboratory Standard Institute (CLSI) guidelines[5] where the antibiotic disks of ceftazidime and ceftazidime-clavulanic acid and cefotaxime and cefotaxime-clavulanic acid were used. A difference of ≥5 mm in the zone diameter of the antibiotic and antibiotic-inhibitor combination indicated positive for the production of ESBL. *E. coli* ATCC 25922 and *K. pneumoniae* ATCC 700603 were used as positive and negative controls respectively for the test.

E. coli strains testing positive for the production of ESBL were analysed for their antibiograms with respect to cephalosporins and other classes of antimicrobials as per CLSI guidelines. Chi-square (χ^2) test was used, a $P \leq 0.05$ was considered to be statistically significant (Epi Info is statistical program available for download online and is distributed by CDC and Prevention, Atlanta).

Results

A total of 363 samples, faces (n = 247) and environmental (n = 116) were collected. *E. coli* strains (n = 361) obtained

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from the different study groups covering humans and environment were analysed for the presence of ESBL production. A total of 124 ESBL producing *E. coli* were recovered. Of the groups studied, the frequency of ESBL producing *E. coli* was found to be 60.3% among the patients, 38% among the healthy individuals and 10.5% among the strains from the environment [Table 1].

The incidence of ESBL producing *E. coli* among the patients was found to be significantly ($P < 0.0001$) higher than the healthy subjects. Among the healthy individuals, 46.1% strains from adults and 25% strains from infants were ESBL producers ($P = 0.001$). The overall frequency of occurrence of ESBL producing *E. coli* was significantly ($P < 0.001$) higher in the human subjects (43.9%) when compared to that of the environment (10.5%). The difference in the incidence of ESBL producing *E. coli* isolated from the hospital environment to that of the other environmental samples however was not statistically significant ($P = 0.092$).

The in-patient sampling included faecal samples mostly from the wards ($n = 64$) and from the Neurology HDU ($n = 13$). From the wards, 83 isolates were recovered, of which ESBL *E. coli* was found to be 60.7%. From the neuro-HDU, 15 isolates were obtained; the frequency of ESBL *E. coli* was 58.3%. The overall resistant rate among the patient group from the wards and the neuro-HDU was 63%. The number of ESBL positives was 43.8% in non-catheterised patients and 16.6% in catheterised patients. The difference among the patients with and without a history of previous hospitalisation was found to be statistically insignificant ($\chi^2 = 1.89$; $P = 0.17$). The data on catheterization, length of hospital stay and previous hospitalisation was not available for eight patients. Out of the 41 ESBL producing *E. coli* reported from the patients group, only one strain, isolated from a patient with septic arthritis was resistant to meropenem and imipenem.

The ESBL producing *E. coli* isolates were resistant to ampicillin and all cephalosporins tested except cefotetan [Figure 1]. The strains recovered from the environment showed very little or no resistance to antimicrobials other than cephalosporins. All the strains showed susceptibility to imipenem, meropenem, tigecycline, amikacin and piperacillin-tazobactam. Only one strain was found to be resistant to imipenem and meropenem. The strains from healthy infants showed more resistance than the strains from the healthy adults.

Discussion

In India, there have been several reports on the prevalence of ESBLs in recent years.[6-9] In our study, of the 363 *E. coli* isolated from different human and environmental sources, 122 were found to produce ESBL. The usage of antimicrobials is one of the major risk factors for colonisation with ESBL producing *Enterobacteriaceae*.[10]

Table 1: Distribution of ESBL *E. coli* among different study groups

Study groups	No. of isolates	No. of *E. coli* (%)	No. of ESBL *E. coli* (%)
Patients (n=77)	98	68 (69.4) (59.7-77.9)	41 (60.3) (48.3-77.4)
Healthy subjects (n=170)	218	189 (86.7) (81.7-90.7)	72 (38.1) (31.4-45.2)
Environment (n=116)	138	104 (75.4) (67.6-82.0)	11 (10.6) (5.7-17.6)

Mid-P value at 95% CI is given in square brackets.
E. coli: Escherichia coli, ESBL: Extended spectrum β-lactamase, CI: Confidence interval

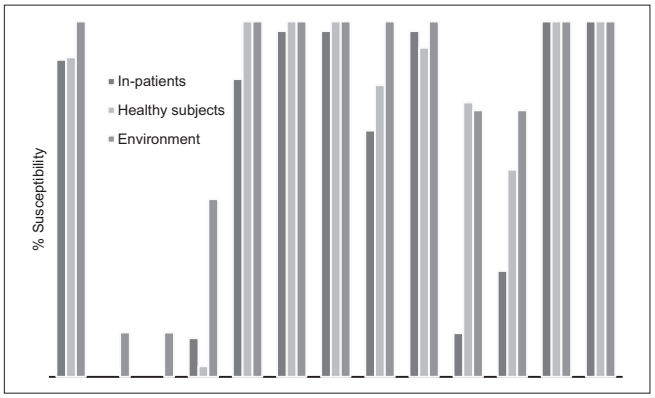

Figure 1: Comparison of antimicrobial resistance pattern of extended spectrum β-lactamases *Escherichia coli* isolated from the study groups

The carrier rate was found to be 25% and 48% among the infants and healthy adults respectively. Several findings suggest that the acquisition of ESBL harbouring isolates may be mediated by contaminated food and water.[11,12] Environment showed a very low rate (10.5%) of ESBL *E. coli*. A high rate of dissemination of *E. coli* in the environment, but not the ESBL *E. coli* indicates a loss of resistance (by losing their plasmids) in the environment. As suggested in a report,[13] for the resistance to succeed, it needs to have a mechanism that imposes fitness burden, along with biologically 'fit' host strain or strains. Therefore, our findings show the environment to be a low risk factor in the acquisition and transmission of ESBL *E. coli*.

ESBL producing strains revealed complete resistance to ampicillin and all cephalosporins used except cefotetan. The strains isolated from the patients showed an increase in resistance rate to other non-β-lactam classes of antibiotics. This could be due to the load of antibiotic pressure in the hospital. When compared with patients, strains from the healthy individuals showed moderate resistance to other classes of antibiotics whereas those from the environment showed very little or no resistance. Carbapenems still stand

as the drug of choice for infections caused by ESBL *E. coli*. Tigecycline showed complete action against ESBL *E. coli* followed by amikacin and piperacillin-tazobactam, only cefotetan showed action against ESBL *E. coli.*

Conclusion

Our study shows high rate of faecal carriage of ESBL producing *E. coli* in patients. The environment was a poor reservoir of ESBL. Our study reinforces the importance of continued surveillance and monitoring of antimicrobial resistance to combat the growing infections caused by ESBL *E. coli*.

References

1. Samaha-Kfoury JN, Araj GF. Recent developments in beta lactamases and extended spectrum beta lactamases. BMJ 2003;327:1209-13.
2. Rossolini GM, D'Andrea MM, Mugnaioli C. The spread of CTX-M-type extended-spectrum b-lactamases. Clin Mirobiol Infect 2008;14:33-41.
3. Pitout JD, Nordmann P, Laupland KB, Poirel L. Emergence of *Enterobacteriaceae* producing extended-spectrum beta-lactamases in the community. J Antimicrob Chemother 2005;56:52-9.
4. Bopp CA, Brenner FW, Fields PI, Wells JG, Strockbine NA. *Escherichia*, *Shigella* and *Salmonella*. In: Murray PR, Baron ES, Pfaller MA, Tenover FC, Yolken RH, editors. Manual of Clinical Microbiology. 8th ed., Ch. 42. Washington, DC: ASM Press; 2003. p. 654-71.
5. Clinical Laboratory Standard Institute. Performance Standards for Antimicrobial Susceptibility Testing: Twentieth Informational Supplement. CLSI Document M100-S20. Wayne: CLSI; 2010.
6. Raghunath D. Emerging antibiotic resistance in bacteria with special reference to India. J Biosci 2008;33:593-603.
7. Ensor VM, Shahid M, Evans JT, Hawkey PM. Occurrence, prevalence and genetic environment of CTX-M beta-lactamases in *Enterobacteriaceae* from Indian hospitals. J Antimicrob Chemother 2006;58:1260-3.
8. Goyal A, Prasad KN, Prasad A, Gupta S, Ghoshal U, Ayyagari A. Extended spectrum beta-lactamases in *Escherichia coli* and *Klebsiella pneumoniae* and associated risk factors. Indian J Med Res 2009;129:695-700.
9. Sankar S, Narayanan H, Kuppanan S, Nandagopal B. Frequency of extended-spectrum β-lactamase producing Gram-negative bacilli in a 200-bed multi-specialty hospital in Vellore district, Tamil Nadu, India. Infection 2012;40:425-9.
10. Gedik H. Does antimicrobial use increase the rate of antimicrobial resistance? A one year experience. Indian J Med Microbiol 2012;30:198-202.
11. Rodríguez-Baño J, López-Cerero L, Navarro MD, Díaz de Alba P, Pascual A. Faecal carriage of extended-spectrum beta-lactamase-producing *Escherichia coli*: Prevalence, risk factors and molecular epidemiology. J Antimicrob Chemother 2008;62:1142-9.
12. Kader AA, Kumar A, Kamath KA. Fecal carriage of extended-spectrum beta-lactamase-producing *Escherichia coli* and *Klebsiella pneumoniae* in patients and asymptomatic healthy individuals. Infect Control Hosp Epidemiol 2007;28:1114-6.
13. Livermore D. The zeitgeist of resistance. J Antimicrob Chemother 2007;60 Suppl 1:i59-61.

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