

Molecular detection of CTX-M-15-type β -lactamases in *Escherichia coli* strains from Senegal

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Abstract

We aimed to detect the extended-spectrum β -lactamases (ESBLs) secreted by clinical strains of *Escherichia coli* at Fann University Hospital in Dakar and to characterize them molecularly. We identified 32 isolates producing ESBLs. The CTX-M-15 gene was the most frequently detected ESBL gene, detected in 90.63% of the isolates studied.

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Keywords: *Escherichia coli*, extended-spectrum β -lactamases, genes, molecular characterization, Senegal

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β -Lactams, the antibiotics most frequently used to treat bacterial infections diseases [1] are becoming less useful against enterobacteria. The principal cause of this decrease in efficacy is the production of extended-spectrum β -lactamases (ESBLs) by these bacteria [1]. In Senegal, only limited molecular characterization data are available for ESBL genes. The overall aim of this study was to detect the ESBLs produced by clinical strains of *Escherichia coli* isolated at Fann University Hospital in Dakar. Our specific objective was to use molecular biology tests to characterize the genes encoding these enzymes.

We studied 32 ESBL-producing isolates of *E. coli* obtained from urine, blood, pus and vaginal secretions. After their isolation and identification between 2009 and 2010, these isolates were stored at -80°C until their molecular characterization. The API 20E panel (bioMérieux, Marcy l'Etoile, France) was used for identification. Antibiogram analyses were carried out by the disc diffusion method on Müller-Hinton agar. ESBLs were detected in tests of synergy between discs carrying third-generation cephalosporins (ceftriaxone, ceftazidime, cefotaxime) and discs carrying amoxicillin–clavulanic acid. The results were interpreted according to the recommendations of the Comité de l'Antibiogramme de la Société Française de Microbiologie (<http://www.sfm-microbiologie.org/>).

Molecular biology tests were carried out at the Bacteriology Laboratory of Pierre et Marie Curie University (Paris VI), France. Total DNA was extracted from the isolates with Qiagen minikit (Qiagen, Germantown, MD, USA). We used forward and reverse primers for the TEM, SHV, CTX-M-9, CTX-M-15, CTX-M-25 and OXA-1 genes [2,3]. PCR products were subjected to electrophoresis in a 3% agarose gel at 100 V for 40 minutes and were purified with the ExoSAP-IT enzyme. Nucleotide sequences were determined by direct Sanger sequencing on an Applied Biosystems 3730 XL sequencer, with the Big Dye Terminator 3.1 cycle sequencing kit (Applied Biosystems, Foster City, CA, USA).

All 32 *E. coli* isolates were susceptible to imipenem. PCR amplification identified the following four genes: TEM, SHV, OXA-1 and CTX-M-15, with a predominance of the CTX-M-15 gene (90.63%; Fig. 1).

To our knowledge, this is one of the first studies to report the isolation of strains of *E. coli* producing CTX-M-15-type ESBLs in Senegal. This type of ESBL was identified in several previous Senegalese studies: a study of 11 enterobacterial isolates, including one *E. coli* isolate [4], and a study of 112 enterobacterial

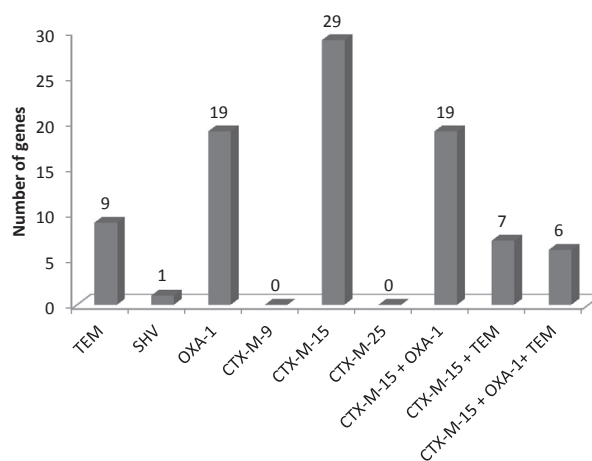


FIG. 1. Distribution of genes found in *Escherichia coli*.

isolates, including 30 *E. coli* [5] isolates, as well as a number of *Salmonella enterica* strains [6]. Our study of 32 isolates of *E. coli*, 90.63% of which harboured the *CTX-M-15* gene, showed that isolates of this type are becoming increasingly common in Senegal, as they are elsewhere in the world [1]. *CTX-M-15*-type ESBLs have also been found in *E. coli* strains in Nigeria [3] and enterobacterial strains in India [7]. The secretion of this enzyme by a strain confers high levels of resistance to cefotaxime, ceftazidime, ceftazidime and aztreonam [8].

This study shows that *E. coli* strains producing *CTX-M-15* ESBLs are circulating in Senegal. Wider surveys are now required to determine the extent to which *CTX-M-15*-producing strains are circulating nationwide.

Conflict of Interest

None declared.

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