IDENTIFICATION OF VARIOUS PATHOTYPES OF DIARRHEAGENIC *E. COLI* USING MULTIPLEX PCR

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Abstract

Background: Diarrheagenic *E. coli* (DEC) are an important cause of childhood and adult diarrhea. DEC are emerging as major contributors of mortality and morbidity especially in children. There is no enough data available regarding the prevalence of DEC in India. Identification of individual DEC pathotypes is based on detection of virulence factors and detection using routine phenotypic laboratory tests is cumbersome as well as time consuming. Identification of DEC is essential for better evidence based management. Here we aimed to study the prevalence of DEC in patients with diarrhea using in-house multiplex PCR assay targeting virulence genes.

Materials and Methods: Consecutive *E. coli* isolates obtained from stool samples of patients with diarrhea during January 2016 to December 2016 were included. DNA was extracted from 100 *E. coli* isolates and respective stool samples using phenol-chloroform method. Multiplex polymerase chain reaction (mPCR) assay was performed using genes encoding the virulence trait of five pathotypes of DEC. The targets selected were *bfpA* for Enteropathogenic *E. coli* (EPEC), vt2 for Enterohemorrhagic *E. coli* (EHEC), *elt B* for Enterotoxigenic *E. coli* (ETEC), *aggR* for Enteroaggregative *E. coli* (EAEC) and *ipaH* for Enteroinvasive *E. coli* (EIEC). mPCR was also performed on direct stool samples using the same protocol. Antibiogram of all the isolates were analyzed.

Results: Out of 100 cases with diarrhea, 24 (24%) cases had DEC infections. EHEC (11%), was the most common DEC followed by EAEC (7%). EIEC and ETEC were found in 3% and 2% of the diarrheal cases respectively. EPEC was detected in only 1% case. Direct PCR on stool samples did not detect DEC from any of the samples.

Conclusion: DEC strains are a significant cause of diarrhea in children as well as adults. The Multiplex PCR assays can be used for simultaneous detection of all DEC pathotypes in routine diagnostic laboratories. These assays are known to be rapid, specific and sensitive. EHEC was predominant pathotype noted in the current study. There was no significant difference between the resistance pattern of DEC and non-DEC. The rate of MDR isolation also was not significantly different in both the groups. The aminoglycoside antibiotics showed dependable sensitivity for both DEC and non-DEC.

Key words: *Diarrhea, E. coli, Multiplex PCR*