

TITLE: Prevalence and detection of virulence factors of enteropathogenic *Escherichia coli* from Brazilian semiarid

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ABSTRACT: Enteropathogenic *Escherichia coli* (EPEC) is one of the most relevant etiologic agents of diarrhea in children. However, molecular epidemiology of EPEC infections are not fully understood worldwide. Several EPEC virulence genes have been described, but their roles on clinical outcomes are not completely defined. In this context, there is a lack of studies that investigate childhood diarrhea in the Brazilian semiarid. This case-control study aimed to determine EPEC prevalence and describe molecular profile of virulence among cases (children with diarrhea) and controls (children without diarrhea) from Brazilian semiarid. A total of 1200 children aging 2-36 months were enrolled from the cities of Crato (CE), Picos (PI), Ouricuri (PE), Cajazeiras (PB), Souza (CE) and Patos (PB). Fecal samples were collected and analyzed for EPEC diagnostic genes (presense of *bfpA* or *eaeA* genes with absence of *stx1* and *stx2* genes) by xMAP technology (Bioplex 200, Biorad). Positive samples were analyzed by Multiplex PCR targeting 19 EPEC virulence factors described in the literature: *espB*, *espD*, *tir*, *espC*, *espZ*, *espL*, *ler*, *map*, *espG*, *espH*, *nleE*, *nleF*, *nleB*, *paa*, *nleC*, *nleD*, *espJ*, *cesT* e *espP*. Among the 1200 children, EPEC was detected in 353 (29.41%). Typical EPEC (positive for *bfpA*) was found in 4.3% of the cases (26/600) and in 1.5% of the controls (9/600) ($p = 0.0053$, OR = 2.974, IC 95% 1.382 to 6.404 by Fisher's exact test), while atypical EPEC (negative for *bfpA*) was detected in 25.3% of the cases (152/600) and in 26.0% of the controls (156/600). Among all EPEC positive samples, *ler*, *cesT* and *espG* genes were the most frequent detected ones (71.3%, 59.2% and 61.1%, respectively), which are associated with global regulation of pathogenicity island. In contrast, the least frequent gene was *nleD*, non-LEE-encoded type III effector. In conclusion, this is the first study to characterize molecular epidemiology of EPEC in the Brazilian semiarid. The findings show significant association of typical EPEC with children with diarrhea and corroborate to higher prevalence of atypical EPEC in the study population. Further investigations are needed for evaluating inflammatory and nutritional status of these children, as well as co-pathogens involved in the outcomes.

Keywords: Enteropathogenic *Escherichia coli*, virulence genes, Multiplex PCR

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