

TITLE: INTERFERENCE OF *Bacteroides thetaiotaomicron* IN THE GROWTH AND VIRULENCE OF ENTEROAGGREGATIVE *Escherichia coli*

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ABSTRACT:

Bacteroides thetaiotaomicron is a member of the microbiota of the intestinal tract of humans. New-found researches revealed its ability to modulate the expression of many virulence genes of enteric pathogens such as *Escherichia coli*. EAEC (Enteraggregative *E. coli*) is an emerging pathogen associated to acute diarrheic infections, persistent diarrhea and chronic intestinal inflammation. The aim of this study was to evaluate the interference of the metabolic products of *B. thetaiotaomicron* in the growth and virulence of EAEC. The growth of EAEC strain cultured in the presence or absence of *B. thetaiotaomicron* supernatant was monitored in anaerobiosis for 9 hours (OD_{600nm}). It was observed that *B. thetaiotaomicron* supernatant increased EAEC's growth. Once adhesion to enterocytes is extremely important for the establishment and persistence of EAEC, qualitative and quantitative adhesion assays of the standard strain were performed on HEp-2 cells and mouse laminin adhesion assays on glass coverslips, in the presence and absence of *B. thetaiotaomicron* supernatant, with initial inoculum of 0.5 (OD_{600nm}) and 5x10⁷, respectively. *B. thetaiotaomicron* supernatant significantly increased adhesion to HEp-2 cells (1,3 x 10¹¹ CFU) when compared to control (4,56 x 10¹⁰ CFU). Laminin adhesion was quantified by capturing 10 random field images from each coverslip [Field of Vision (FV) = 0.30mm²]. EAEC adhesion to laminin was improved by *B. thetaiotaomicron* supernatant (604.3 CFU / FV) in comparison to the strain cultured with medium (500.73 CFU / FV). Both conditions were compared with their respective controls (BSA 2%), quantified at 49.11 CFU / FV and 96.58 CFU / FV, respectively. Biofilm production was also investigated in both culture conditions of the standard and clinical EAEC strains in which most strains were characterized as strong (> 1) and moderate (0.79) biofilm producers in the presence of *B. thetaiotaomicron* supernatant. Motility tests were also performed from culturing strains with *B. thetaiotaomicron* supernatant, in which the motility was increased about 6-fold. These results allow us to infer that the *B. thetaiotaomicron* supernatant is capable of significantly interfering with the main virulence factors of EAEC. Further studies are required to characterize specific metabolites to elucidate the interspecies interactions of pathogens and commensals in the intestinal environment, which may be involved in the persistence and / or virulent behavior of this species.

Keywords: *Bacteroides thetaiotaomicron*; *Escherichia coli*; Pathotypes; Microbiota; Metabolome

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