Title: Molecular characterization of *Shigella sonnei* strains isolated in the São Paulo State during 21 years.

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Abstract

*Shigella sonnei* is an important causative agent of human diarrheal diseases worldwide, mainly in children under five years. In Brazil, *S. sonnei* is not usually studied, therefore studies that molecularly characterize strains of this species are of great importance. The aims of this study were to genotype by Pulsed-field gel electrophoresis (PFGE) and investigate the frequency of virulence-associated genes by Polymerase chain reaction (PCR) in *S. sonnei* strains isolated from sick humans in this country. A total of 58 *S. sonnei* strains isolated from human diarrheic feces between 1983 and 2004 in São Paulo State were studied. For PFGE, genomic DNA was digested with 40U of *Xba*I and the fragments resolved in a CHEF-DRIII apparatus. Data were analyzed by BioNumerics 5.1 version software and the dendrogram constructed by UPGMA method and DICE similarity coefficient. All the 58 strains studied presented the genes *ipaH, iuc* and *sigA*. The *ipaBCD* gene was found in 13 (22%) strains. The genes *ial* and *virF* were detected in 12 (20%) strains. The *sen* gene was found in 6 (10%) strains. The genes *set1A, set1B, sat, pic* and *sepA* were not detected in any strains. The dendrogram of genetic similarity of PFGE grouped the strains in two clusters, designated A and B. The cluster A contained 35 strains isolated between 1983-2004 with 33 strains with a similarity above 80.3%. The cluster B contained 23 strains isolated between 1984-2002 with 20 strains with a similarity above 81.6%. Among clusters A and B strains the similarity was below 70.6%. The Discrimination index (DI) of PFGE was 0.996. The pathogenic potential of the strains studied was highlighted by the presence of important virulence-associated genes searched. The PFGE results may suggest that two prevalent subtypes that differed little genotypically prevailed and contaminated humans during 21 years in the important metropolitan region of Ribeirão Preto in the São Paulo State.

Keywords: *Shigella sonnei*, virulence genes, PFGE, molecular typing.

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