

TITLE: Distribution of the *pilS* gene encoding the pilin of the type IV pilus, Pil, among typical enteropathogenic *Escherichia coli* strains

AUTHORS: CASTRO, F.S.¹; GARCIA, B.G.¹; VIEIRA, M.A.M.¹; GIRÃO, D.M.², DOS SANTOS, L.F.³, PIAZZA, R. M. F.⁴, HERNANDES, R.T.⁵, GOMES, T.A.T.¹.

INSTITUTION: UNIFESP - Universidade Federal de São Paulo, Disciplina de Microbiologia, São Paulo, SP, Brasil¹

Universidade Federal do Rio de Janeiro, Instituto de Microbiologia Prof. Paulo de Góes, Rio de Janeiro, RJ – Brasil²

Instituto Adolfo Lutz, Divisão Técnica de Biologia Médica. São Paulo, SP, Brasil³

Instituto Butantan, Laboratório de Bacteriologia, São Paulo, SP, Brasil⁴

Universidade Estadual Paulista, Instituto de Biociências. Botucatu, SP, Brasil⁵

ABSTRACT: Typical enteropathogenic *Escherichia coli* (tEPEC) are important diarrheal agents in children in the first year of life. They are characterized by the ability to establish a localized adherence (LA) pattern and to form attaching and effacing lesions in cultured epithelial cells. Another *E. coli* pathotype, enteroaggregative *E. coli* (EAEC), is characterized by the formation of the aggregative adherence (AA) pattern in HeLa cells and biofilm formation. Recently, we reported the occurrence of EPEC strains of serotype O119:H6 producing the LA and an AA-like pattern concurrently (LA/AA-like+). These strains carried a large plasmid containing the *pil operon* that encodes Pil, a type IV fimbriae that is responsible for the AA pattern production in some EAEC strains. In this study, we investigated the prevalence of two alleles of *pilS* (encoding the structural pilin subunits) among tEPEC strains of different serotypes, geographic locations and years of isolation. One hundred and eighty nine strains (33 different serotypes), which were isolated from diarrheic and non-diarrheic children and lacked all five EAEC Aggregative Adherence Fimbriae (AAF)-encoding genes were tested by PCR to detect the *pilS* gene. Two pairs of primers were used, which were designed based on the sequences of the *pil operon* of the EAEC C1096 strain (*pilS*_{C1096}) and of the LA/AA-like+ tEPEC EC404/03 strain from our collection (accession number AP014805.1). Overall, the *pilS* gene was found in 87 (46%) of the strains, none of which carried both alleles. In addition, *pilS*_{C1096} occurred most often than *pilS*_{Ec404} (25.4% e 20.6%, respectively). Of the total of 87 positive strains, 25 (28.7%) belonged to serotype O111:H2 (20 *pilS*_{C1096} and 5 *pilS*_{Ec404}) and 24 (27.6%), to O119:H6 (24 *pilS*_{Ec404} and 4 *pilS*_{C1096}). Among the tEPEC serotypes, we have found that *pilS*_{C1096} were more prevalent in the O111:H2 and *pilS*_{Ec404} were more prevalent in the O119:H6 (22.9% and 27.6%, respectively). Interestingly, up to the 1990s, these serotypes were the most frequent among tEPEC strains of diarrheic infants in São Paulo. The adherence patterns of the strains as well as the involvement of Pil in the establishment of the AA-like pattern among these strains are under investigation. The presence of additional colonization attributes might improve the virulence of tEPEC strains.

Keywords: enteropathogenic *Escherichia coli*, hybrid adherence pattern, localized adherence, Aggregative adherence, Pil fimbriae.

Development Agency: Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) e Fundação de Amparo à Pesquisa do Estado de São Paulo