TITLE: SAT (SECRETED AUTOTRANSPORTER TOXIN) IS INVOLVED IN IMMUNE SYSTEM EVASION OF *ESCHERICHIA COLI* ISOLATED FROM BACTEREMIA

AUTHORS: CLAUDIA A. FREIRE, ROSA M. SILVA, ANGELA S. BARBOSA, WALDIR P. ELIAS

INSTITUTIONS: LABORATÓRIO DE BACTERIOLOGIA, INSTITUTO BUTANTAN, SÃO PAULO, SP, BRAZIL; DISCIPLINA DE MICROBIOLOGIA, ESCOLA PAULISTA DE MEDICINA, UNIVERSIDADE FEDERAL DE SÃO PAULO, SÃO PAULO, SP, BRAZIL.

Extraintestinal *Escherichia coli* (ExPEC) are the main Gram-negative pathogens associated with bacteremia and sepsis. The capacity of those strains in reaching the bloodstream relies on a diverse set of virulence factors (VFs), and those involved in immune system evasion are especially important to establish this kind of infection. Serine Protease Autotransporters of Enterobacteriaceae (SPATE) are secreted virulence factors involved in cytotoxicity and immune modulation, including degradation of complement system molecules. In a previous study, our group searched for SPATE-encoding genes in a collection of *E. coli* strains isolated from bacteremia, detecting *sat* (secreted autotransporter toxin) as the most frequent gene. Sat is a cytotoxin originally described in uropathogenic *E. coli* and further found in other *E. coli* pathotypes. Considering that other SPATE members described as cytotoxic proteases are also capable of causing immunomodulatory effects, such as the cleavage of the complement molecules, this study aimed to investigate a possible role of Sat in serum resistance. For this purpose, one of those *sat*-positive *E. coli* (EC-071) was selected for whole-genome sequencing, using the Illumina HiSeq 1500 platform, and *in silico* prediction of known VFs, using the ecoli VF collection database. The VFs analysis confirmed the presence of *sat*. Moreover, no known serum resistance-encoding genes were detected. Although, EC-071 was able to survive in a serum bactericidal assay when incubated with 50% normal human serum. Sat was then purified from EC-071 concentrated supernatant in an anion exchange column and its identity confirmed by mass spectrometry. Purified Sat (5 µg) was incubated with C4 or C4b complement molecules during 5 or 24 h, and the products of the incubations were analyzed by immunodetection with anti-C4 antibodies. Degradation of both C4 and C4b was observed in both incubation times. Our results show that Sat, a frequent SPATE found in *E. coli* strains isolated from bacteremia, plays an important role in evasion of the host immune system.

Keywords: ExPEC, bacteremia, SPATE, Sat.

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