# The type III secreted effector EspZ of enteropathogenic E. coli protects against lytic host cell death by targeting ABCB6

**B. Aroeti**[I], G. Szakacs[II], M.M. Gottesman[III], E. Rivkin-Zlotkin[I], S. Banerjee[I]

[I]Department of Biological Chemistry, Alexander Silberman Institute of Life Sciences, The Hebrew University of Jerusalem, Jerusalem, Israel, [II]Center for Cancer Research, Medical University of Vienna, Spitalgasse 23, Vienna A-1090, Austria, [III]Laboratory of Cell Biology, Center for Cancer Research, National Cancer Institute, National Institutes of Health, 37 Convent Drive, Room 2108, Bethesda, MD 20892, United States of America

Enteropathogenic E. coli (EPEC) is a significant human bacterial pathogen that adheres to intestinal epithelial cells, causing severe infectious diarrhea (gastroenteritis), mainly in children in developing countries. EPEC virulence depends on a type III secretion system (T3SS), a molecular syringe the pathogen utilizes to inject (translocate) a series of bacterial proteins, termed effectors, into host cells. These effectors target various host cell proteins and lipids to subvert vital processes for the pathogen's benefit. However, the mechanisms underlying the effectors' activities are not fully understood. The translocated intimin receptor (Tir) is the first and most abundant effector injected into the host cell plasma membrane. Tir is crucial for infection because it provides a receptor for intimate bacterial attachment to the host cell surface. Upon translocation, Tir triggers the formation of filamentous (F)-actin-rich plasma membrane protrusions, called pedestals, on top of which the bacterium resides. Tir has also been implicated in triggering lytic host cell death, inhibiting bacterial colonization. However, the second translocated essential effector, EspZ, immediately counteracts this activity. The mechanism by which EspZ blocks Tir-induced lytic cell death is unknown. Both Tir and EspZ are necessary for eliciting the EPEC disease. Using co-precipitation and proteomics, we identified the ATP binding cassette subfamily B member 6, ABCB6, as a primary host binding partner of EspZ. Interestingly, confocal imaging revealed that ABCB6 (full length and core) colocalizes with EspZ at F-actin-rich pedestals at infection sites. Importantly, knocking out ABCB6 expression by CRISPR/Cas9 reduced the ability of injected EspZ to protect against lytic host cell death and localize at infection sites. These results argue that EspZ trafficking, positioning, and host cell death protective activity are promoted by its ability to interact with ABCB6.