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Quantifying the Protein-Protein Interaction between the Ion-Channel-Forming Colicin A and the Tol Proteins by Potassium Efflux in E. coli Cells

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Abstract : Colicins are a family of bacterial toxins that kill Escherichia coli and other closely related species. The mode of action of colicins involves binding to an outer membrane receptor and translocation across the cell envelope, leading to cytotoxicity through specific targets. The mechanism of colicin cytotoxicity includes a non-specific endonuclease activity or depolarization of the cytoplasmic membrane by pore-forming activity. For Group A colicins, translocation requires an interaction between the N-terminal domain of the colicin and a series of membrane- bound and periplasmic proteins known as the Tol system (TolB, TolR, TolA, TolQ, and Pal and the active domain must be translocated through the outer membranes. Protein-protein interactions are intrinsic to virtually every cellular process. The transient protein-protein interactions of the colicin include the interaction with much more complicated assemblies during colicin translocation across the cellular membrane to its target. The potassium release assay detects variation in the K+ content of bacterial cells (K+in). This assays is used to measure the effect of pore-forming colicins such as ColA on an indicator organism by measuring the changes of the K+ concentration in the external medium (K+out) that are caused by cell killing with a K+ selective electrode. One of the goals of this work is to employ a quantifiable in-vivo method to spot which Tol protein are more implicated in the interaction with colicin A as it is translocated to its target.

Keywords : K+ efflux, Colicin A, Tol-proteins, E. coli
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