Adhesin Degradation Accelerates Delivery of Heat-labile Toxin by Enterotoxigenic *Escherichia coli*

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Tips for understanding the paper:

serine protease - protein which cleaves other proteins at a serine residue

type V (autotransporter) secretion system

a self-contained protein translocation system ... a secretion system which transports proteins across both the inner and outer membranes of the Gram (-) cell

The secreted protein has the following domains:



The N-terminal signal peptide targets the protein to a transporter complex (Sec) in the inner membrane which transports it across to the periplasm. In the process this signal is cleaved from the protein.

The β -barrel region inserts into the outer membrane, forming a pore.

The passenger domain of the protein slides through the pore to the outside of the cell. It is released from the cell by autocleavage at the junction between the passenger domain and the β -barrel domain.



Heat-labile toxin (LT)

an enterotoxin (protein) produced by ETEC (see MoMD page 201 and fig 16-2) identical to cholera toxin You should read MoMD page 132 ("structure-function organization of exotoxins"), page 133 ("cholera toxin"), and figure 9-3

Caco-2 cells

see Wikipedia article http://en.wikipedia.org/wiki/Caco-2

bacterial adhesin

see MoMD page 117 ("surface colonization")

experimental nomenclature

"r" in front of the gene (as in rEatA) stands for "recombinant"

"p" after the name (as in rEatAp) indicates the passenger domain (a <u>protein</u>, not the gene)

rEtpA.myc.His

a recombinant protein, produced by fusing the EtpA gene to gene sequences encoding a short domain from the myc gene, followed by a region encoding 6 consecutive histidines

The resulting fusion protein contains three domains:

- a. EtpA
- b. 10 amino acids of the myc protein sequence (a protein "tag)
- c. 6 histidine residues (a protein "tag")

This is useful because the protein can be purified using immobilized antibodies against the myc tag or the polyhistidine tag. Alternatively the protein can be purified by chromatography on a nickel column, to which the polyhistidines bind.

Figure 3d

This is a "far Western" blot, used to detect protein-protein interaction. Read the description in the "Experimental procedures." You may also read more about this method by looking it up online.

If you do not understand the meaning of a word, LOOK IT UP! If you do not understand a concept or technique, LOOK IT UP!

This is what scientists do. 🙂