Crystal Structure of the Ferric Enterobactin Receptor from *E. coli*

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Ferric enterobactin receptor (FepA) is an 80 kD integral outer membrane protein from *E. coli* that functions to bind ferric enterobactin and transport it into the periplasm; it also serves as a receptor for colicins B and D. FepA belongs to the family of high-affinity, active-transport outer membrane receptors that specifically transport iron and vitamin B_{12} across the outer membrane. All members of this family derive energy for transport from the proton motive force across the inner membrane; this is accomplished by contacting an inner membrane protein, TonB. By analogy with bacterial porins, FepA is expected to use amphipathic beta strands to span the membrane. However, with a molecular weight of 80 kD, the predicted beta barrel and corresponding pore should be considerably larger than observed for the known structures of diffusion-controlled porins.

FepA has been overexpressed in the outer membrane of *E. coli*. Isolation and purification in the presence of detergent yields protein which retains its specificity and binding affinity for ferric enterobactin. Crystals suitable for diffraction experiments have been grown from solutions containing detergent and polyethylene glycol 1000.[1] The crystals have the symmetry of the space group C222, with a = 112.3 Å, b = 127.6 Å, and c = 135.7 Å. The crystals diffract to at least 2.5 Å resolution and there is one monomer in the asymmetric unit. Initial phases were obtained by MIRAS from site-directed cysteine mutants; the structure was subsequently solved using MAD data obtained from a selenomethionyl crystal at -170˚C. The structure exhibits a monomeric beta barrel which utilizes contacts between extracellular loops of one monomer and periplasmic loops of an adjacent monomer to pack into a type II membrane protein crystal. Details of the topology, ferric enterobactin recognition, and unpredicted features will be presented. Elements of the structure relating to active transport will be discussed.

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[1]Smith, B. S., Kobe, B., Kurumbail, R., Buchanan, S. K., Venkatramani, L., van der Helm, D., and Deisenhofer, J. (1998). "Crystallization and Preliminary X-ray Analysis of Ferric Enterobactin Receptor FepA, an Integral Membrane Protein from *Escherichia coli. Acta Cryst. D*", in press.