Expression of E. coli capsular polysaccharide requires the KfiB protein:
A Structural based analysis

Abstract

**Background and objectives:** Capsular polysaccharide expression is an important virulence factor for many invasive bacterial pathogens of humans. Escherichia coli offer a model system to study the mechanisms by which capsular polysaccharides are synthesized and exported onto the cell surface of bacteria. Biosynthesis of the E. coli K5 capsular polysaccharide, which consists of the repeat structure -(1, 4)-GlcA-[α(1, 4)]-GlcNAc-[α(1-, requires the KfiA, KfiB, KfiC, and KfiD proteins. Until now, the role of all proteins except KfiB have been known.

**Material and Methods:** To study the role of the KfiB protein, a full-length (pMA1) and several derivatives of KfiB expression construct (pMA2-6) were made with a 6Xhis-tag at the N-terminus. The presence of the hexa histidin tag facilitated purification of the KfiB protein using Ni²⁺-NTA chromatography.

**Results:** All plasmids except pMA1 failed to complement the mutation in pPC6::23 and restore capsule production. Successful complementation by pMA1, showed that the fused hexa-histidine tag did not interrupt the function of KfiB and that the full-length KfiB is required for complementation.

**Conclusion:** Localization studies revealed that KfiB is associated with the cytoplasmic membrane.

**Key words:** Escherichia coli, Capsular polysaccharide, KfiB, K5