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

Pourhossein, M.(PhD)
Assistant Professor of Molecular Biology,Isfahan university of Medical Sciences

Roberts,I.S(PhD)
Professor, Smith Building, Oxford Road, Manchester University, Manchester, UK

Corresponding Author:
Pourhossein M

E.mail:
pourhossein@med.mui.ac.ir

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 -4) GlcA-β(1, 4)-GlcNAc-α(1-, requires the KfiA, KfiB, KfiC, and KfiD proteins. Until now , the role of all proteins except KfiB have been kown.

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 Ni^{2+}-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