

ABSTRACT

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Title of diploma thesis: Carbohydrate recognition of the novel colonization factor CS30 of enterotoxigenic *E. coli*

Enterotoxigenic *Escherichia coli* (ETEC) is a major cause of diarrhoea in developing countries and travellers to these areas, as well as in many farm animals. The main ETEC virulence factors are two enterotoxins and outer membrane proteins called colonization factors that mediate the adherence of the bacteria to the host epithelial cell surface in the small intestine. Carbohydrate recognition of the recently identified colonization factor CS30 was characterised using binding studies with the mixture of glycosphingolipids isolated from the human and porcine intestine. The CS30 binding compound was identified as a mixture of sulfatides with different ceramide species by mass spectrometry and a binding assay with monoclonal antibodies directed towards SO₃-3Galβ. Further testing confirmed that CS30 binds to the glycosphingolipid with a terminal SO₃-3Galβ and prefers sulfatides with the longer fatty acid chain. To prevent false positive results in the binding studies, a recombinant strain TOP10-CS30 containing CS30 operon that harbours seven genes *csmA-G* was constructed. RT-PCR confirmed transcription of all seven genes encoding the CS30 fimbriae, but transmission electron microscopic imaging showed no fimbrial structures on the cell surface, suggesting that the building of the fimbriae requires native regulatory genes.

Keywords: ETEC, diarrhoea, CS30, carbohydrate recognition, sulfatide, TOP10-CS30