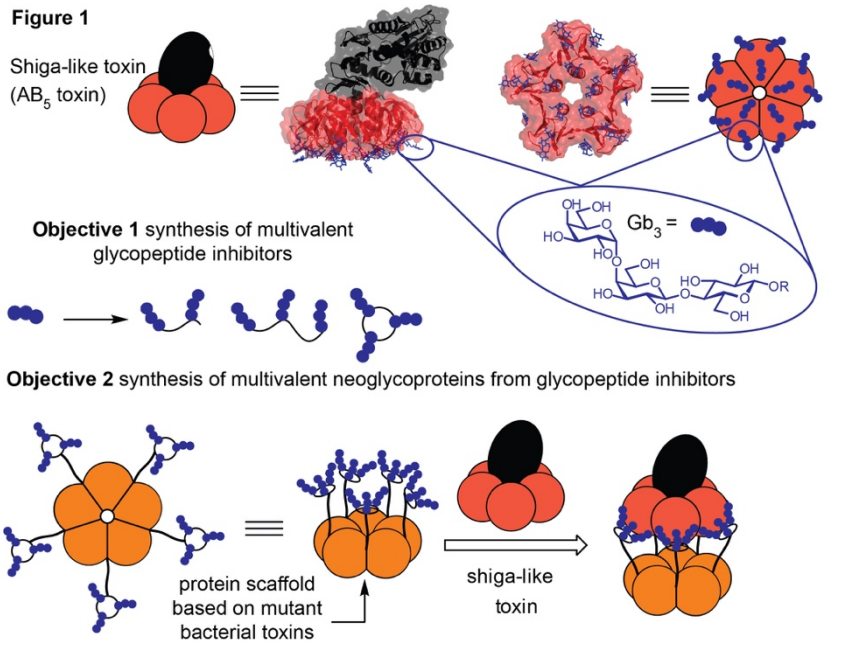
GlypStx
Glycoprotein-based inhibitors of shiga-like toxin

Gastrointestinal infections have substantial impact in both the developing world and in Europe with 1.5 billion cases each year leading to approximately two million deaths each year, 760,000 of which are in children under five years old. Many of these deaths are caused by bacteria for example E. coli O157, that produces protein toxins, including shiga-like toxin. In the human body, the surfaces of living cells are covered in complex carbohydrate molecules. This "sugar coating" allows the cells to interact with viruses, bacteria, and toxins that have complementary protein receptors. The shiga-like toxin (Fig. 1), produced by *E. coli* O157 bacteria, is part of a family of AB_5 toxins which have a single toxic A protein that is associated with a pentagonal sugar-binding B protein that delivers the toxin into the cells that line the intestine. The B protein can stick to fifteen copies of a specific lipid-linked carbohydrate called Gb3 on the gut wall. The result is bloody diarrhoea and the toxin entering the circulatory system to cause kidney failure. Each individual interaction is very weak, but by working together, many weak interactions give a very strong adhesion, just like the many loops and hooks in Velcro. These so-called "multivalent interactions" are very common in the biology of carbohydrates.

In this project, we are mimicking these natural multivalent interactions to make inhibitors that can stop the toxin from sticking to, and entering cells. Our inhibitors are based on artificial glycoproteins (neoglycoproteins) that have five or fifteen copies of the target sugar attached at specific sites on a protein scaffold that has the same size and shape as the target toxin (Fig. 1). The aim is that they can bind to all five of the toxin's receptor sites simultaneously providing an extremely strong interaction to prevent the toxin from entering cells. During the project we have synthesised a series of glycopeptides and attached them to pentameric protein scaffolds using chemistry that allows very precise modifications of the protein scaffolds. The optimized inhibitors will have potential as a new class of future biopharmaceuticals.

Figure 1



This project has been the work of Dr Vajinder Kumar who has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement no. 746421

UNIVERSITY OF LEEDS