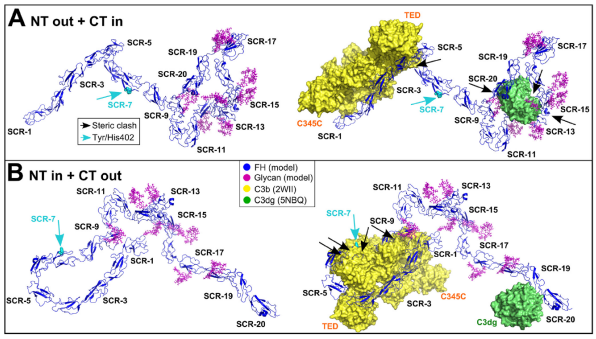
Principal Investigator: Prof S J Perkins

Grant Title: CCP-SAS: solution structure modelling of important glycans and oligosaccharides

ABSTRACT: OBJECTIVES. The field of macromolecular crystallography has resulted in over 150,000 biological crystal structures, one reason for this being the existence of appropriate software, such as the CCP4 computational crystallography initiative. The equivalent software revolution is now needed to advance research in glycans and oligosaccharides through understanding their atomistic structures in solution. The availability of modern small-angle X-ray and neutron scattering (SAXS, SANS) instruments at large multiuser facilities provides this solution structural data for novel biological science applications. Thus we need to integrate experimental data on glycans with user-friendly, high-throughput, molecular modelling of the data.

METHODS: CCP-SAS (Collaborative Computational Project in Small Angle Scattering) was set up in 2012 by ~10 groups from the UK and the USA. In 2016, CCP-SAS released a web-accessible portal called SASSIE-web that is a complete but simple workflow. This runs molecular dynamics and Monte Carlo simulations on a high performance computing (HPC) platform to model proteins and nucleic acids and fit these to experimental scattering data. This Mizutani proposal was aimed to fill an important research gap in relation to solution structures for carbohydrates on proteins.

RESULTS: With the awarded funds, we completed and published a full modelling analysis of a glycoprotein called complement factor H that contains eight N-linked glycan moieties (total molecular mass 155 kDa) in the 20 small “short complement regulator” (SCR) domains of Factor H (see Figure). The attachment of the glycan chains to the protein proved challenging, as well as assembling the initial full molecular model of Factor H from its 20 domain structures. The improved accuracy of the models intriguingly suggested that Factor H exhibited two different glycosylated structures from the best fits (see Figure). One structure fitted well with the binding of the major activated complement C3b to the N-terminus of Factor H (see A in the Figure). The other structure fitted well with the binding of the fragmentation product C3d to the C-terminus of Factor H (see Figure B). Experiments are now in progress to test the existence of these two conformers. Other projects worked on include heavily glycosylated antibody structures, such as IgE and IgA1. IgE was modelled with 12 N-linked glycan chains, while IgA1 has three O-linked and two N-linked glycans. The outcomes of these modellings showed that our procedure is effective and produces biologically-interesting structures. We thus showed that CCP-SAS has the potential to transform carbohydrate science, and to dramatically accelerate the discovery process with these important macromolecules.



A, B. Two Factor H conformations