Synchrotron radiation circular dichroism (SRCD) spectroscopy investigations of the structure and orientation of membrane proteins in lipid bilayers

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Membrane proteins are physiologically vital, accounting for up to 60% of pharmacological targets, but notoriously challenging to purify and characterise. Methods to investigate their structure are often laborious, requiring microgram quantities of protein. Most approaches do not provide the lipid bilayer milieu that is well established as significant in both the structure and function of membrane proteins.

Oriented SRCD spectroscopy is a powerful and sensitive tool to investigate the structure of membrane proteins in lipid bilayers (mimicking native cellular membranes). Using the B23 beamtime at Diamond Light Source we have been developing methods for the preparation and measurement of oriented SRCD samples to characterise the structure of integral membrane proteins and their orientation with respect to the lipid bilayer. In the work presented here we outline methods for the preparation of oriented lipid bilayers using mechanical and magnetic alignment, and present initial oriented SRCD spectra and correlating solid-state nuclear magnetic resonance (NMR) data.

As a model membrane protein for these measurements we are using the transmembrane domain (TMD) of the putative glycosyltransferase Fukutin-1 (Fk1), whose mislocalisation has been implicated in the onset of Fukuyama congenital muscular dystrophy. The localisation of Fk1 in the late endoplasmic reticulum (ER)/Golgi apparatus is thought to be mediated through changes in oligomeric state and structure induced by the surrounding lipids. Using oriented samples for both SRCD and solid-state NMR we are beginning to understand how the distinct lipid composition within the late ER/Golgi may govern the retention of this protein in these compartments.