

**P019** Ligand binding and activation of chemokine receptor CXCR6

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The chemokine CXCL16 is found in both soluble and membrane-bound forms and is a ligand for the GPCR known as CXCR6. As a membrane-anchored chemokine, CXCL16 supports cell adhesion and in its soluble form it induces cell migration. We are studying the CXCL16: CXCR6 interaction at the molecular level. Chimeric CXCR6: CXCR1 constructs were generated, exchanging the N-termini of the receptors. Unlike cells expressing wild-type receptor, both mutants were inactive in chemotaxis and adhesion assays, despite efficient cell surface expression. This suggests that binding of CXCL16 and subsequent activation requires an intact CXCR6 N-terminus.

Since previous work has shown basic residues of CXCL16 to be important for CXCR6 binding a panel of 17 CXCR6 point mutants were generated to probe ligand binding, cell adhesion and chemotaxis. Whilst the single point mutation D176 was found to perturb CXCL16 binding, mutation of E274 within the seventh helix of CXCR6 resulted in an inability to bind and migrate in response to soluble CXCL16 whilst leaving adhesion to membrane-anchored CXCL16 unimpaired. Thus, we hypothesize that E274 of CXCR6 is a key residue in the activation of CXCR6 by ligand and that its mutation forces the receptor to adopt a conformation unable to interact with soluble CXCL16.