Synthesis of a PAR2-Targeting Peptide Library with Biased Signaling Properties

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Protease-activated receptor 2 (PAR2) is constitutively expressed on the endothelial cells of blood vessels, playing a role in numerous physiological processes such as cell migration, vasodilation, and inflammation. Upon PAR2 activation, several second messenger signaling cascades are activated to regulate these cellular functions. Studies have found that certain ligands can promote selective activation of one or more signalling pathways, a concept referred to as biased agonism. However, it is unclear which PAR2-mediated functions are coupled to each signalling pathway in endothelial cells. This work aims to design PAR2 activating peptides that produce endothelium-dependent vasodilation with little to no inflammation.

A library of 25 PAR2-targeted seven-mer peptides was synthesized by automated Fmoc solid-phase peptide synthesis, purified by preparative HPLC, and characterized by high-resolution mass spectrometry. Modifications to the C-terminal region explored the importance of different structural features on receptor activation. Initial results revealed peptides that were biased towards specific G protein pathways when containing a positively charged C-terminus, and other pathways when containing short aliphatic or polar uncharged amino acids at position 6. These structural features will also be evaluated for functional selectivity, providing insight into how the different PAR2 driven functional responses are coupled to each signalling pathway.