TOWARDS UNDERSTANDING G PROTEIN-COUPLED RECEPTOR: A MULTIDISCIPLINARY APPROACH

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G protein-coupled receptors (GPCRs) constitute the largest family of cell surface receptors. There are 800-1000 genes in the human genome which encode for these receptors. Despite large variations in their stimuli, all GPCRs share a common seven transmembrane helix architecture and perform signal transduction by a common mechanism via heterotrimeric guanyl nucleotide binding proteins (G proteins). GPCRs play regulatory roles in many different physiological processes and they represent one of the most important class of drug targets. Due to the lack of three-dimensional structures, structure based drug design has not been possible. During this study, selected GPCRs were produced and characterized in different heterologous expression hosts. Subsequently, the receptors were purified using affinity chromatography, which yielded milligram amounts of pure and stable receptors. Purified receptors were used for three dimensional crystallization trials and solid state NMR analysis to obtain strutural information on these proteins. Using solid state NMR, we have successfully obtained the the receptor bound conformation (i.e. active conformation) of bradykinin (a peptide ligand for the human bradykinin receptor). This information should facilitae design of potent and specific drugs acting on the bradykinin receptor. Additionally, it was also found that co-expression of the human angiotensin receptor is required to promote surface expression and functional response of the human bradykinin receptor. The mechanism underlying the heterodimerization of these two receptors is being investigated further.