Agonist interactions and selectivity in $GABA_{A/C}$ receptors

B.A. Cromer, H.S. Tae and S. Petrou, Health Innovations Research Institute, School of Medical Sciences, RMIT University, Bundoora, VIC 3083, Australia and Howard Florey Institute, University of Melbourne, Parkville, VIC 3095, Australia.

Cys-loop ligand-gated ion channels constitute one of two major superfamilies of receptors mediating rapid chemical synaptic transmission in the central nervous system. They include cation selective channels that are receptors for excitatory neurotransmitters, acetylcholine and serotonin, and anion selective channels that are receptors for inhibitory neurotransmitters, γ-aminobutyric acid (GABA) and glycine. Recent structural information from snail acetylcholine binding proteins (AChBP), torpedo acetylcholine receptors and bacterial homologs have provided a good understanding of the overall structure of the superfamily and of specific details of acetycholine-receptor interactions. For inhibitory receptors for GABA and glycine, however, we have a much more limited understanding of how receptors interact with and are selectively activated by particular agonists.

In this study, we have investigated interactions between GABA and receptor, using the homopentameric ρ1 γ-aminobutyric acid receptor ($GABA_C$) as a model for the broader family of heteropentameric $GABA_A$ receptors. We used homology modeling to identify a series of conserved charged residues at the GABA-binding site that we hypothesized formed a series of charge-charge interactions likely to be important for interaction with agonist, agonist selectivity and receptor activation. We have tested this hypothesis using site-directed mutagenesis in combination with two-electrode voltage clamp recording of recombinant receptors expressed in *Xenopus* oocytes. Preliminary results have revealed key determinants of agonist selectivity, particularly determining sensitivity to the size or length of the ligand, as well as receptor activation or gating. These results are consistent with our hypothesis and provide a basis for a more detailed understanding of agonist-receptor interactions in inhibitory Cys-loop ligand-gated ion channels.