Centre for Integrative Physiology

Research Briefings

Molecular pharmacology and novel signalling pathways of G protein-coupled receptors (GPCRs)

G PROTEIN-COUPLED RECEPTORS (GPCRS), INCLUDING THOSE RESPONDING TO DOPAMINE OR 5-HYDROXYTRYPTAMINE (5-HT), REPRESENT KEY THERAPEUTIC TARGETS FOR THE TREATMENT OF PSYCHIATRIC DISORDERS, SUCH AS SCHIZOPHRENIA AND DEPRESSION. WE ARE INVESTIGATING THEIR PROFILE OF INTRACELLULAR SIGNALLING PATHWAYS, DIFFERENTIAL INVOLVEMENT OF THESE IN EVENTS ASSOCIATED WITH PSYCHIATRIC DISORDERS, GENETICALLY DETERMINED DIFFERENCES AND THE POTENTIAL OF NEWER THERAPEUTIC AGENTS TO ENGAGE SPECIFIC SIGNALLING PATHWAYS FROM SUCH RECEPTORS.

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INTRODUCTION

While many psychiatric disorders probably arise through a complex interplay of genetic traits and experience-mediated events, which may cause abnormal neurodevelopmental progression, effective therapeutic interventions are centred on GPCRs such as those for dopamine or 5-hydroxytryptamine. These receptors act to modulate forebrain activity underpinning psychiatric function. 5-HT2A receptors for example are a primary target of leading atypical antipsychotic agents and we are working to elucidate how clinically useful drugs differentially engage with different signalling pathways of these receptors.

NEW GPCR SIGNALLING PATHWAYS

Many GPCRs can activate phospholipase D (PLD) to generate phosphatidic acid. This is an important second messenger, which regulates a number of downstream signalling cascades including ERK MAPkinase and plays key roles in vesicular trafficking, subcellular recruitment of signalling proteins and cellular growth/proliferation.

We discovered a novel enhanced linkage to PLD activation in particular GPCRs that requires a specific sequence motif at the cytosolic face of the receptors, carrying out site-directed mutagenesis to achieve appropriate gain- or loss-of-function. These receptors' facilitated route to PLD signalling involves recruitment, direct binding and activation of small monomeric G proteins such as ARF and Rho, as opposed to the classical heterotrimeric G proteins. Ongoing work is addressing the dynamic operation and regulation of this linkage in relation to alternative or concurrent signalling events.

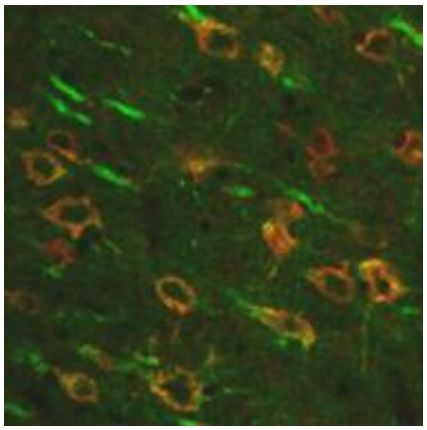

Figure 1: Immunofluorescence imaging of pyramidal neurones in frontal cortex showing co-localisation of the 5-HT2A receptor (green) with an orphan GPCR (red), which is implicated in psychiatric disorders by molecular genetic studies but whose endogenous ligand is not yet known.

NEW GPCR PARTNERS

Certain GPCRs can link to a variety of partner proteins, which modify their properties and intracellular signalling profiles. Examples can be seen at the level of receptor heterodimerisation and in the interactions with

chaperone, scaffolding or explicit signalling proteins.

We are working on new intracellular partners of the 5-HT2A receptor (which strongly activates PLD signalling) both by exploring predicted candidates and by targeted proteomics. The work has revealed a novel enhanced signalling complex incorporating the small G protein ARF1 and phospholipase D1 assembled around specific motifs in the carboxy-terminal tail of the receptor. Understanding how this pathway operates in the face of recruitment and dissociation of other receptor partners, heterotrimeric G proteins and arrestins is a key goal.

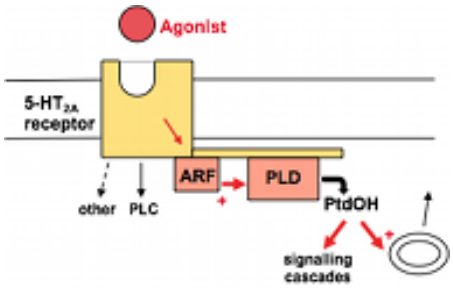

Figure 2: The 5-HT2A receptor, a target for newer antipsychotic agents, displays a novel pathway of phospholipase D (PLD) activation through a molecular complex in which the small G protein, ARF1 and PLD1 assemble around different sequence motifs in the tail of the receptor. The PLD product, phosphatidic acid (PtdOH) regulates vesicular trafficking and a range of downstream signaling cascades. Conventional signalling of the receptor is through phospholipase C (PLC) and other pathways.

PHARMACOLOGICALLY AND GENETICALLY DETERMINED SELECTIVE SIGNALLING

Increasing evidence points to an exciting new era of molecular pharmacology, ligand-induced selective signalling, in which it is becoming possible to identify agonists that dock to receptors in a way that causes preferential activation of one signalling pathway over another. We are actively investigating this in the context of small G protein-dependent PLD signalling compared to conventional signalling by the 5-HT2A receptor as it is an important target of psychotherapeutic agents. We recently discovered that a single nucleotide polymorphism in the 5-HT2A receptor gene that is associated with poor response to newer antipsychotic agents results in selective disruption of ARF/PLD signalling by the receptor while conventional signalling is largely maintained.

ADDITIONAL ACTIVITIES

In ongoing collaboration with Sue Fleetwood-Walker's group we are pursuing a range of projects addressing molecular mechanisms of pain and analgesia. This work has led to an active drug discovery programme focused on mechanistically novel analgesics and to the translational application of some of our key findings in both clinical and veterinary settings.

Selected references:

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