Developing a new and improved version of the operational model of pharmacological agonism

Background

Within the field of G protein-coupled receptor (GPCRs) pharmacology, it has now become apparent that, rather than single signalling moieties, GPCRs are pleotropic, activating multiple intracellular pathways – this concept is know as biased agonism. How we quantitate bias has been something that has been the focus of a great deal of research. Initial attempts relied upon researchers comparing the maximal effect (E_{max}) and the potencies (EC_{50}) of agonists for different pathways. However, we now appreciate that these methods fail to account for spare receptors or the implicit differences observed in amplification of different assays. For example, in second-messenger assays (which have significant amplification) both full and partial agonists may reach E_{max} whereas in an assay with little amplification (e.g. monitoring recruitment of β-arrestin to a receptor by enzyme complementation) partial agonists exhibit significantly lower maximal effects. To address these issues a number of analytical methods have been developed that build on the operational model of agonism proposed by Black and Leff in 1983.

The operational model of pharmacological agonism
The operational model of pharmacological agonism is probably one of the first examples of systems pharmacology - attempting to separate key drug specific parameters (binding affinity - K_A) from system parameters (resulting activity - τ) that includes both receptor density and coupling efficiency. By normalising the transducer ratio (τ/K_A) for a test ligand against a reference ligand, at a reference pathway, we obtain a quantity referred to as the bias factor that is proposed to negate the impact of cell background and assay condition. Unfortunately however, despite its wide spread use, the model has many flaws. Thus we feel it is timely and of significant importance to try to update or improve on the operational model to account for these multiple flaws.

Specific questions we would like to answer
1. Can we devolve a generic models that accounts for multiple paths of signalling that result in positive or negative signalling effects dependent on the agonist concentration?
2. How can we account for receptor internalisation, recycling to cell surface nor degradation and re-synthesis when modelling GPCR pathway bias data/
3. Can we account for the important role of the G protein involved or other regulatory molecules in modelling agonist bias?
4. Can the new model be extended to account for allosteric modulation without adding to much complexity?
5. Can we account for constitutive activity of the receptors in our new model?
6. Can the new model account for the temporal dynamics of the assays being used?

Biological data available
The laboratory of Dr Graham Ladds (Cambridge) has a wealth of pathway specific bias data from multiple GPCRs that can be modelled. Many GPCRs are pleotropic resulting in changes in the intracellular levels of cAMP, Ca^{2+}, pERK1/2 activity, pAKT1/2/3 activity and even cell survival. All these pathways can be interlinked or be activated separately. For example, pERK1/2 activation can occur via elevation of cAMP or through recruitment of β-arrestin proteins to the activated GPCR. Further many GPCRs also undergo endo/exocytosis processes that are influenced by the intracellular machinery and the

activating agonist. We will provide data for two GPCRs, glucagon-like peptide 1 receptor and the adenosine A1 receptor for which we have quantitated rates of second messenger production, pERK1/2 activation and receptor internalisation. Finally, we also have data available where we have quantitated the kinetics of agonist binding to both GPCRs.

References

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