[P17] Discrimination of G–protein coupled receptors and their conformational states using intramolecular interaction

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G–protein coupled receptors (GPCR) are membrane receptors able to transmit stimuli to cells. The molecular mechanism of signal transmission involves the receptor coupling to effector in response to ligand binding, and this depends on the receptor conformational state. To date, the 3D-structures of 50 different GPCRs have been characterized by X-ray crystallography and the dynamics of some of them have been extensively studied by molecular dynamics simulation, suggesting general mechanism of activation/inactivation.[1]

Here we propose a new method to compare different GPCR structures, independently of predefined structural or functional determinants. This method is based on the detection and comparison of intramolecular non-covalent interactions in the seven transmembrane domains (TM).

In more details, the analysis of a 3D–structure involves the extraction of TM coordinates followed by the representation of hydrogen bonds (labeled with *inter–helix* or *intra–helix*, and with *with sidechain* or *within backbone*), ionic bonds and aromatic bonds as either a graph or a fingerprint built from Ballesteros-Weinstein numbering.[2] Comparing two 3D–structures does not require that they are described in a common frame. Two graphs are aligned for the bestfit superimposition of the maximum common substructure. Similarity between two fingerprints is calculated using the Tanimoto coefficient.

We have applied the method to the classification of 215 GPCR structures available in the Protein DataBank. Networks built from the comparison of graphs showed that *with sidechain inter/intra–helix* hydrogen bonds are sufficient to differentiate GPCRs. All polar interactions except *within backbone intra–helix* hydrogen bonds well differentiate the activation states of a GPCR. Global analysis of interactions suggested specific signatures of GPCRs and their activation state.

We have also applied the method to the analysis of two molecular dynamics trajectories where a GPCR experiences a transition from the active to the inactive state[3,4] The all-against all comparison of frames delimited a few clusters. The characterization of clusters by consensus interaction fingerprints revealed which interactions are state–specific.

In conclusion, we developed a new method able to discriminate GPCR from a simplified 3D-representation (8-46 interaction points). The same approach also distinguishes conformational states and has proved to successfully cluster and describe the conformational states generated by molecular dynamics simulation.

Bibliography:

[1] Manglik, A.; Kobilka, B. The Role of Protein Dynamics in GPCR Function: Insights from the B2AR and Rhodopsin. *Curr. Opin. Cell Biol.* **2014**, *27*, 136–143.
[2] Desaphy, J.; Raimbaud, E.; Ducrot, P.; Rognan, D. Encoding Protein–Ligand Interaction Patterns in Fingerprints and Graphs. *J. Chem. Inf. Model.* **2013**, *53* (3), 623–637.
[3] Miao, Y.; McCammon, J. A. Graded Activation and Free Energy Landscapes of a Muscarinic G-Protein–coupled Receptor. *Proc. Natl. Acad. Sci.* **2016**, *113* (43), 12162–12167.
[4] Dror, R. O.; Arlow, D. H.; Maragakis, P.; Mildorf, T. J.; Pan, A. C.; Xu, H.; Borhani, D. W.; Shaw, D. E. Activation Mechanism of the B2-Adrenergic Receptor. *Proc. Natl. Acad. Sci.* **2011**, *108* (46), 18684–18689.