COLLOQUIA IN CELLULAR SIGNALLING

Venue: Medical University Vienna, Center for Physiology and Pharmacology,
Institute of Pharmacology, Waehringerstrasse 13a, 1090 Vienna, "**Leseraum**".
(Harald Sitte, Tel.: (01) 40160 31323, *harald.sitte@meduniwien.ac.at*

Thursday **13.02.2014 11:00 s.t.** **Jean-Philippe Pin (host: H. Sitte)**

University of Montpellier
CNRS, INSERM
Institute of Functional Genomics
F-34094 Montpellier

"Metabotropic glutamate receptors: complex allosteric machines for fine-tuning of synaptic activity"

Jean-Philippe Pin (jean-philippe.pin@igf.cnrs.fr)

Abstract:
Metabotropic glutamate (mGlu) receptors are key modulators of synaptic transmission, regulating both pre- and post-synaptic processes. As such they are considered as new targets for drug development for both psychiatric and neurologic diseases. These receptors are more complex proteins than any other GPCRs, being composed of two subunits, each composed of three main domains: a venus flytrap domain (VFT, where glutamate binds) connected to a heptahelical domain (7TM, where positive allosteric modulators (PAM) bind) through a cystein-rich domain (CRD). Such complexity opens a number of possibilities for drug development, but raised a number of issues on how the receptor is activated and regulated.
Using a combination of molecular and biophysical tools, we show that their dimeric organization is essential for agonist activation. Indeed, agonist binding in the cleft of each VFT help stabilize a new orientation of the two VFTs, leading to a relative movement of the two 7TM domains. Using single molecule analysis, we demonstrated that the VFT dimer oscillates between a resting and an active state on a sub-millisecond timescale, and that ligands exclusively influence the transition rates between these conformations.
When analyzed at the level of the 7TM domains, we confirmed that a single 7TM domain is activated per dimer, as a result of the reorientation of the two 7TMs within the dimer. We identified TM4-5 as the main interface in the inactive state, while TM6 is part of the active dimer interface. Of note, locking the TM4-5 interface with a disulfide bond prevent receptor activation, while locking the TM6 interface generates a constitutively active receptor.
Taken together, these data illustrate the coordinated allosteric transitions that link agonist binding into the VFT clef, to G protein activation in such dimeric GPCR complexes, and provide important information for our understanding of the asymmetric activation mode of dimeric GPCRs.

1- El Moustaine et al, *PNAS* 2012; 2- Hlavackova et al, *Science Sig.* 2012; 3- Doumazane et al, *PNAS* 2013.