Master de Sciences et Technologies
Mention Biologie Intégrative et Physiologie
Parcours : Neurosciences
Responsable : Professeur Régis Lambert

Internship Proposal
Academic Year 2019-2020

1. Host team :

Research Unit (e.g. Department or Institute) : UMR8246, Neuroscience Paris Seine
Research Unit Director : Hervé Chneiweiss
Research Team Director : Bertrand Lambolez and Bruno Cauli
Team name : Synaptic and Neuroenergetic Networks

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2. Internship project title: *Signaling and interactions in the mGluR1/5-GluD1 receptor complex*

3. Internship Description :

Glutamate is the main excitatory neurotransmitter in the brain. It binds ionotopic receptors (iGlu) and metabotropic receptor (mGlu). Among iGlus, GluD1 and GluD2 subunits are peculiar as they do not directly bind glutamate. Mutations in the *GRID1* gene coding for GluD1 have been identified in patients suffering schizophrenia or autistic disorders. The phenotype of GluD1 knock out (KO) mice resembles that of patients. Interestingly, GluD subunits are implicated in the development and maturation of synapses due to their interactions with pre-synaptic released cerebellins (Cbln). Our team has recently demonstrated that GluD1 is widely expressed in the brain and forms a signaling complex with mGlu1/5 receptors and that activation of mGlu1/5 induces the opening of the GluD1 channel. Our collaborators have identified a rare missense mutation in *GRID1* in patients with intellectual disability (ID). We showed that this mutation ($GluD1^{ID}$) does not alter membrane expression or the interaction of GluD1 with mGlus. However, when overexpressed in primary neuronal cultures, $GluD1^{ID}$ leads to a reduced number and length of neurites and an increase of the number of immature spines. We have also shown that $GluD1^{ID}$ disturbs mGlu1/5 mediated ERK signaling, a pathway strongly involved in synapse formation and plasticity. These results prompted us to generate a mouse model carrying the human mutation to investigate the role of GluD1 and $GluD1^{ID}$ in synaptogenesis and synaptic transmission.

With this new model we will

1) Image mGluR1/5 mediated intracellular pathways (ERK, Calcium, PKA) in neurons in brain slices using fluorescent biosensors
2) investigate the effect of the mutation on the synaptic activity and plasticity using electrophysiology

Depending on the progress of the project, the student will be involved in one or more of the following aspects:
• Regulation of mGlu5 signaling by wild type and mutant GluD1. The activation of intracellular signaling pathways will be investigated using western blotting and biosensor imaging.
• Study of the binding of Cbln to mutant GluD1 using immunocytochemistry, in cell western blotting and/or surface plasmon resonance.
• Analysis of the GluD1 interactome: Wild type and mutant GluD1 binding partners will be identified using immunoprecipitation and mass spectrometry. Binding partners will be validated using transfected HEK cells and functional significance evaluated accordingly.

GluD1 and type I mGlus are key player in synaptopathies associated diseases but the molecular mechanism linking GluD1 mutations to mental disorders remains largely unknown. This proposal aims to understand how mutant GluD1 leads to synaptic deficits causing ID. Identification of new GluD1 interacting partners and disturbances caused by the mutation will help understand the pathophysiological mechanisms linking GluD1 to synapse formation, function and plasticity. Elucidation of the molecular cascade that links GluD1 to ID has enormous potential to identify new therapeutic targets relevant ID models.

IMPORTANT NOTE: GluD1 being an iGlu, alteration of the channel properties by the mutation will also be examined using patch-clamp recordings. This technique is widely used in the laboratory. Students motivated by this aspect of the project are welcome to postulate.