PROPOSAL ABSTRACT:

Name of Principal Investigator:	**Pablo R. Moya**
Proposal Title:	**Molecular and Functional Evaluation of a Mouse Model of Obsessive-Compulsive Disorder with Increased EAAT3 Expression.**

Obsessive-Compulsive Disorder (OCD) is a severe neuropsychiatric disorder affecting 2-3% of the population worldwide. The overarching characteristics of OCD are intrusive, usually senseless thoughts (obsessions) and repetitive, intentional behaviors (compulsions). OCD symptoms are partially responsive to standard treatments, being substantially impairing and persistent through lifetime (Pauls et al., 2014). The only well-studied medications are the selective serotonin reuptake inhibitors (SSRIs) that block the serotonin transporter; however, 40-60% of patients are unresponsive to SSRIs (Murphy and Moya, 2011). Although there is a clear need for novel and better therapies for OCD, there are no novel medications approved since early 1990s. Therefore, it is imperative to better understand the neurobiological basis of OCD to find novel targets that might lead to improved pharmacological treatment for this devastating disorder. Accumulating evidence from genetics, neuroimaging and animal model studies indicate alterations in the glutamatergic system in OCD. From a genetics perspective, the *SLC1A1* gene, encoding the neuronal glutamate transporter EAAT3, is the most consistent candidate gene for OCD. EAAT3 is highly expressed in brain regions implicated in OCD (Ahmari, 2016). In a previous grant, we found that mice with increased EAAT3 expression in forebrain using CaMKIIa driver (EAAT3glo/CMKII) display increased anxiety-like behavior, increased repetitive behavior and increased grooming, all behaviors relevant to OCD phenotype and that were alleviated upon chronic, but not acute (S)SRI treatment. Thus, we demonstrate that increased EAAT3 expression contributes to OCD pathophysiology (Delgado et al., under review in *Neuropsychopharmacology*, NPP-18-0863).

Highlight findings from our past grant indicate that, in EAAT3glo/CMKII mice have altered glutamate NMDA receptor subunit composition and impaired NMDA-dependent synaptic plasticity at the corticostriatal synapses, a key component of the CSTC circuit. It is known that NMDAR subunit composition changes from high NR2B-containing to high NR2A-containing NMDARs during synapse maturation and in response to activity and experience (Paoletti et al., 2013). Our finding of increased corticostriatal NR2B-containing NMDARs in adult EAAT3glo/CMKII mice raises the possibility that the NR2A/NR2B developmental switch is impaired in this model; this will be evaluated in this new proposal in several brain areas relevant to OCD. Besides to the original goals from our previous grant, we also found that EAAT3glo/CMKII mice display increased spontaneous recovery of fear memory. This remarkable finding is consistent with the pathophysiology of OCD and agrees with prior reports of impaired retention of extinction in human OCD patients and animal models of OCD (Milad et al., 2013, Reimer et al., 2017). Since deficits in extinction are associated with impaired ventromedial prefrontal cortex (vmPFC) (Lüthi & Lüscher, 2014), our findings suggest impairments in PFC functionality in EAAT3glo/CMKII mice; this will be also evaluated in this proposal.

This proposal is aimed to gain a deeper insight on the molecular and functional changes underlying the behaviors relevant to OCD in EAAT3glo/CMKII mice. Our **working hypothesis** is **"glutamatergic alterations in the striatal and prefrontal areas in EAAT3glo/CMKII mouse lead to synaptic and circuit impairments underlying the OCD relevant behaviors".** We will evaluate developmental changes in cell morphology and in the composition and function of glutamate NMDA and AMPA receptors in several brain regions involved in anxiety, repetitive behaviors and recall of extinction; we expect changes in NR2A/NR2B ratio at the synapse reflecting the functional changes found in electro-physiological recordings. We will test if synaptic changes found in corticostriatal synapses also occur in PFC, which is implicated in extinction deficits and compulsivity. To elucidate neural circuit correlates of behaviors relevant to OCD, we will perform *in vivo* electrophysiological simultaneous recordings in brain regions involved in anxiety, compulsivity and extinction (orbitofrontal cortex, vmPFC, dorsomedial striatum, amygdala) of behaving EAAT3glo/CMKII mice. We expect to find differences in temporal firing patterns and oscillatory power in at specific frequencies, and to find sets of neurons showing activity changes during specific behaviors relevant to OCD. Finally, we will evaluate the potential anti-compulsive profile of a novel, non-commercial selective EAAT3 blocker. **This would allow us to strengthen our EAAT3glo/CMKII model for neuropsychiatric research and to firmly establish EAAT3 as a novel target for OCD**. The present proposal takes advantage of the expertise of our collaborative group in behavioral neuroscience, molecular biology, *in vitro* and *in vivo* electrophysiology to generate an interdisciplinary approach contributing to understand the role of novel candidate genes in animal models of neuropsychiatric disorders.