Abstract

The recent discovery of the Ca^{2+}/calmodulin dependent protein kinase II alpha subunit (CaMKIIα) hub domain as the γ-hydroxybutyric (GHB) high affinity binding site provided for novel potential therapeutic applications in neuroscience of GHB analogs. CaMKIIα belongs to the family of CaMKII proteins and it is involved in the regulation of calcium signaling in the CNS, which is underlying neuronal plasticity and physiological processes such as learning and memory consolidation. CaMKIIα is also implicated in neurodegenerative diseases and ischemic stroke. The deep cavity of the CaMKIIα hub domain remains still unexplored despite the availability of a variety of GHB analogs, among which HOCPCA and Ph-HTBA represent promising neuroprotective agents.

The work discussed in this PhD thesis is directed at the investigation on multiple fronts of the CaMKIIα hub domain using different approaches and expanding the chemical scope to elucidate the structure and the function of the latter.

In *Chapter 3*, fluorinated derivatives of the brain permeable NCS-382 scaffold have been selected for the development of the first PET tracers to image the CaMKIIα hub domain. Organic synthesis has been optimized and radiochemistry experiments are ongoing.

In *Chapter 4* the CaMKIIα hub domain was further explored since an additional cavity has been identified within the domain. This new binding pocket could represent a new druggable target which will allow the modulation of the primary binding site to achieve more selective and efficient ligands. Derivatives of the GHB analogs O-5-HDC and PTCA have been employed for the development of potential bitopic ligands together with PAM fragments. Two compounds showed additional interactions, however the bitopic nature has not been confirmed yet.

In *Chapter 5* the in-house GHB analog PTCA has been further investigated. To improve the brain exposure of PTCA, biological cleavable esters were synthesized. Also, considering the need of additional BBB permeable GHB high affinity ligands, modifications and substitution of the thiazole core were performed. Two compounds showed high cellular permeability and have been excluded as efflux transporter substrates nor BCRP substrates. Preclinical PK studies are ongoing to assess in vivo brain penetration.