OC-02. DESIGN AND PRECLINICAL DEVELOMENT OF CP86, A NEW POTENT IN VIVO ANTIEPILEPTIC AGENT

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Kv7.2/7.3 potassium channels represent an attractive pharmacological targets for the treatment of different neurological disorders, in particular epilepsy. During 2011 the Kv7.2/7.3 agonist retigabine has been approved as add-on treatment for drug-resistant partial onset seizures with or without secondary generalization. However, the clinical use of retigabine has been declining over the years, leading to its market withdrawal in 2017,[1] due to several drawbacks and side effects. The resulting limitation in therapeutic options led to extensive efforts, both from academia and industry, in the search for new Kv7.2/7.3 agonists. These efforts were largely frustrated by a substantial lack of knowledge concerning retigabine binding site.[1] This is why, starting from our previously published results,[1] we have performed in-silico studies to shed light on the chemical space at the retigabine binding site. A structure-based approach was used to verify the in-silico hypotheses synthesizing a focused library of 25 compounds, which were tested by an HTS fluorometric assay. Results obtained further expanded the structure-activity relationship clues for the rational design of Kv7.2/7.3 channels agonists, confirming the pivotal importance of a wide, lipophilic pocket in correspondence of the pore region for the modulation of Kv7.2/7.3 agonists potency and efficacy. Nevertheless, as assessed by photostability testing, synthesized compounds were unable to overcome one of the main retigabine drawback: light-induced instability. Thus, taking advantage form the previously obtained SAR clues, a photostability-driven design was performed, generating a second set of 16 molecules. Among these, CP86 showed improved potency, efficacy and photostability when compared to retigabine. Patch-clamp experiments confirmed these preliminary data. CP86 is able to produce a marked leftward shift of the dose response curve and increased maximal currents in comparison to Kv7.2/7.3 prototypical agonists. Site-specific mutagenesis experiments validated the predicted binding mode, involving an interaction network with W236, V225, F240, S303, F304, F305 and L312, in the Kv7.2/7.3 subtype heterotetrameric assembly. CP86 was, then, subjected to an extensive in vitro and in vivo preclinical characterization resulting as a metabolically stable compound, with considerably improved half-life and CNS distribution if compared to retigabine. When challenged in vivo, by a pentylenetetrazol (PTZ) kindling model of epilepsy, CP86 showed a remarkable reduction of the incidence and severity of tonic PTZ seizures, at one-twelfth of the retigabine dose. Moreover, seizures modulation was combined with an outstanding protective effect. CP86 administration, indeed, largely prevented pentylenetetrazol-induced death, widely described[2] and experimentally observed for retigabine-treated animals.

References:

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