Pharmacology of AMPA receptors expressed in cell lines and stem cell-derived neurons

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The vast majority of excitatory neurotransmission is mediated by AMPA (a-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid) receptors. The functional receptor exists as a tetramer, either homomers or heteromers, from a repertoire of 4 different subunits, GluA1 – GluA4. It is well known that glutamate is a neurotoxin and it is proposed that overactivation of ionotropic glutamate receptors may underlie many neurodegenerative disorders such as ischemic stroke, epilepsy, Parkinson's and dementia, amongst others. Enhancement of AMPA receptor activation by, for example, BDNF, has been proposed to have beneficial effects of learning and memory and has potential therapeutic value in the treatment of depression, Huntington's and Parkinson's diseases. We have used GluA2 receptors expressed in HEK cells on 3 different automated patch clamp systems recording from either 1, 8 or 384 cells simultaneously. We have compared the glutamate EC_{50} obtained on the different platforms and found that they are similar, ranging from approximately 30 – 60 µM. Responses to glutamate were reproducible using a concentration of 30 or 100 µM, GluA2 could be repetitively activated at least 3 or more times, making the assay suitable for pharmacological characterization. Using pharmacological agents we could either inhibit or enhance the glutamate elicited responses. The inhibitor, CNQX, blocked glutamate responses mediated by GluA2 with an IC_{50} of 600 nM using a glutamate concentration of 100 µM and the concentration response curve for CNQX was dependent on the glutamate concentration. The potentiator, LY404187, was pre-incubated prior to co-application with 100 µM glutamate giving an EC_{50} of around 500 nM.

In addition to GluA2 expressed in HEK cells, we also recorded glutamatergic-enriched cortical neurons derived from induced pluripotent stem cells on an automated patch clamp platform. In these neurons we recorded NaV currents which could be blocked by low concentrations of TTX with an IC_{50} of 12 nM. In addition, glutamate responses were recorded which were potentiated by LY404187.