021P mGLUR4A INHIBITS N AND P/Q-TYPE CALCIUM CHANNELS TO DEPRESS EVOKED GLUTAMATE RELEASE IN THE RAT ENTORHINAL CORTEX

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We have previously demonstrated that activation of presynaptic group III metabotropic glutamate receptors (mGluR) enhances the spontaneous release of glutamate at synapses in layer V of the entorhinal cortex (EC), evidenced by an increase in spontaneous excitatory postsynaptic current (sEPSC) frequency. This effect was due to activation of the mGluR4a subtype (Evans *et al*., 2000a). However, EPSCs evoked by concurrent stimulation of afferent inputs (eEPSCs) were concurrently depressed (Evans *et al*., 2000b). In the current study we have determined whether the effect of mGluR4a activation on eEPSCs is mediated by inhibition of voltage gated Ca^{2+}-channels (VGCC).

EC slices were prepared from the male Wistar rats (50-110g). Whole cell voltage clamp recordings were made from neurones in layer V, visualized using differential interference contrast optics and an infrared video camera. eEPSCs were elicited by electrical stimulation (10-100 mV, 0.02 ms duration) via a bipolar electrode placed on the surface of the slice in layer V lateral to the recording site. A paired Students t-test was used for comparison of eEPSC amplitudes. All error values refer to standard error of the mean. Drugs were applied by bath perfusion.

The specific mGluR4a agonist, ACPT-1 reduced the amplitude of eEPSCs from 29.4±1.2 pA to 18.0±1.4 pA ($P<0.05$). Specific inhibitors were then used to determine whether inhibition of VGCC may underlie this effect. ω-conotoxin GVIA (CTx, 400 nM) was used to block N-type channels and agatoxin IVA (AgTX, 200 nM) and Ni^{2+} (25 μM) to block, P/Q and R-type channels, respectively. CTx reduced the normalized mean fractional eEPSCs to 0.75±0.02 ($P<0.01$; n=8). A similar effect was seen with AgTx (0.75±0.02; $P<0.01$; n=8). The Addition of ACPT-1 with either toxin saw further reductions to 0.49±0.02 and 0.52±0.03, respectively ($P<0.01$in both cases). Ni^{2+} reduced the fractional eEPSC amplitude to 0.71±0.03 ($P<0.01$; n=7), but in the presence of ACPT-1 the additional reduction, to 0.26±0.01, was much more pronounced ($P<0.01$).

Thus, N, P/Q and R-type channels all contribute to glutamate release at these terminals. Inhibition of either P/Q or N-type channels alone did not prevent the depression of the eEPSC by mGluR4a activation, but blockade of R-type channels essentially doubled it. One explanation for these data would be that the effect of ACPT-1 depends on inhibition of both N and P/Q-type, but not R-type channels. Inhibition of either P/Q or N-type channels alone would allow the unblocked N or P/Q-type channels to fully mediate the effects of mGluR activation. In these circumstances, mGluR-insensitive R-type channels would help maintain eEPSC amplitude. With R-type channels blocked, glutamate release would depend only on N and P/Q-type channels, both of which would be inhibited by mGluR activation, strongly reducing the intraterminal Ca^{2+} transient and hence the eEPSC.

Evans, D I P *et al*., (2000a). J. Neurophysiol. 83, 2519-2525.

Evans, D I P *et al*., (2000b). J. Physiol. 527P, 100P.