Steady State Expression of L-Type Calcium Channels in the Superior Region of the Rat Hippocampus

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Studies have shown that the metabotropic receptor for γ-aminobutyric acid, $GABA_BR$, works through second messenger systems to regulate the mammalian nervous system. Studies in our lab have shown that $GABA_B$ receptors mediate facilitation of voltage dependent L-type calcium channel current in a subset of hippocampal neurons. This facilitation has only been seen in early postnatal rat pups, with the facilitation peaking in the second postnatal week. Studies in the laboratory suggest that $GABA_B$ receptors mediate this facilitation through activation of protein kinase C (PKC). These observations suggest that PKC, once activated by $GABA_B$ receptors, may phosphorylate select L-type channels to produce the facilitation in calcium current. Of the four different high voltage activated L-type channels, only two are expressed in the rat hippocampus, $Ca_v1.2$ and $Ca_v1.3$. Nuñez and McCarthy (Dev. Neurobio, 67: 1879-1890, 2007) have demonstrated that expression of the cardiac L-type calcium channel ($Ca_v1.2$) peaks between birth and 14 days in rats. Thus, we hypothesized that specific L-type calcium channels may be expressed at different time points in the first two postnatal weeks and that this could explain why facilitation is primarily present in the second postnatal week. The current study examined the steady state expression of the L-type calcium channel proteins, $Ca_V1.2$ and $Ca_V1.3$, using Western blot analysis. Hippocampal proteins were extracted from the superior region of 1 to 42 day old rat pups and separated by SDS-PAGE. For analysis the proteins were transferred to PVDF membranes and were decorated with antibodies specific for $Ca_v1.2$ and $Ca_v1.3$, as well as β-actin to control for loading. The integrated optical density (IOD) of the bands decorated with the calcium channel antibodies were normalized by dividing them by the IOD of the bands labeled with β-actin antibodies. $Ca_V1.3$ demonstrated an increase in steady state expression throughout development, thus decreasing the likely hood of its role in the L-type current facilitation through the $GABA_B$ receptor activation. Alternatively, $Ca_V1.2$ steady state expression peaked during the first two postnatal weeks. This expression pattern suggests that $Ca_V1.2$ may play a role in the facilitation of L-type calcium current by $GABA_B$ receptor activation during the early postnatal period.