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The effects of 1,25-MARRS silencing on cabindin D28k expression in primary cortical neurons

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The limited studies have indicated that beta amyloid cytotoxicity can lead depletion of calcium binding proteins such as calbindin in neuronal cells [1, 2]. Vitamin D regulates the expression of calbindin D28k in several cell types [3]. Our previous study has showed that amyloid β (Aβ) treatment eliminated VDR protein in cortical neurons [4]. These results might indicate the potential role of vitamin D and vitamin D mediated mechanisms in neurodegeneration [5]. However there was no data about the regulation of calbindin D28k under the condition of 1,25-MARRS repression. The aim of this study was to investigate the expression levels of calbindin D28k in 1,25-MARRS silenced primary cortical neurons.

Cortical neuron cultures were prepared from Sprague-Dawley rat embryos (Figure 1). 1.25-MARRS was silenced with siRNAs. qRT-PCR was performed for determining the expressions of calbindin D28k. Immunofluorescent labelling was performed for determining the localization of calbindin D28k protein.

Our findings showed that the mRNA levels of calbindin D28k did not change in cortical neurons in response to 1,25-MARRS down-regulation ($p>0.05$). Immunoreactivity for Calbindin D28k was peripherally localized in cytoplasm of cortical neurons (Figure 2).

Our results indicated that the disruption of vitamin D-1,25-MARRS pathway in our model do not exert any effect on calbindin D28k expression.

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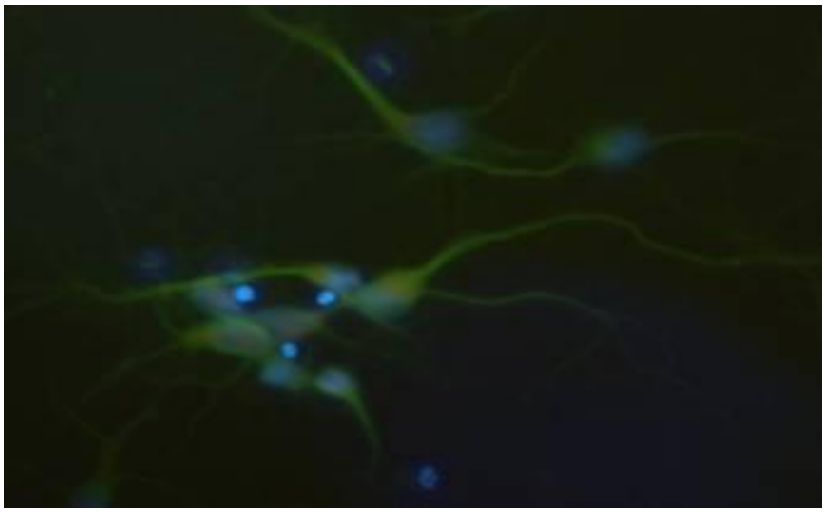

Figure 1. 7 days old primary cortical neuron culture. Neurons (green; FITC tagged Pan Neuronal Marker antibody), nuclei (blue; DAPI), x40.

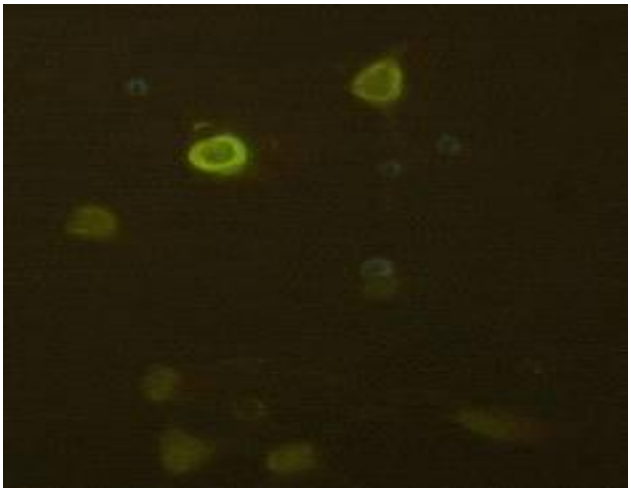

Figure 2. 8 day old primary cortical neurons. Immunoreactivity for Calbindin D28k is peripherally localized. FITC tagged anti-Calbindin D28k antibody. X40