The L-type Ca\textsuperscript{2+} channels blocker nifedipine represses mesodermal fate determination in murine embryonic stem cells.

Abstract:
Dihydropyridines (DHP), which nifedipine is a member of, preferentially block Ca(2+) channels of different cell types. Moreover, influx of Ca(2+) through L-type Ca(2+) channels (LTCCs) activates Ca(2+) signaling pathways, which in turn contribute to numerous cellular processes. Although LTCCs are expressed in undifferentiated cells, very little is known about its contributions to the transcriptional regulation of mesodermal and cardiac genes. This study aimed to examine the contribution of LTCCs and the effect of nifedipine on the commitment of pluripotent stem cells toward the cardiac lineage in vitro. The murine embryonic stem (ES, cell line D3) and induced pluripotent stem (iPS, cell clone 09) cells were differentiated into enhanced green fluorescence protein (EGFP) expressing spontaneously beating cardiomyocytes (CMs). Early treatment of differentiating cells with 10 µM nifedipine led to a significant inhibition of the cardiac mesoderm formation and cardiac lineage commitment as revealed by gene regulation analysis. This was accompanied by the inhibition of spontaneously occurring Ca(2+) transient and reduction of LTCCs current density (I(CaL)) of differentiated CMs. In addition, nifedipine treatment instigated a
pronounced delay of the spontaneous beating embryoid body (EB) and led to a poor surface localization of L-type Ca(2+) channel ?(1C) (Ca(V)1.2) subunits. Contrary late incubation of pluripotent stem cells with nifedipine was without any impact on the differentiation process and did not affect the derived CMs function. Our data indicate that nifedipine blocks the determined path of pluripotent stem cells to cardiomyogenesis by inhibition of mesodermal commitment at early stages of differentiation, thus the proper upkeep Ca(2+) concentration and pathways are essentially required for cardiac gene expression, differentiation and function.