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Expression of potassoim channel in stem cell derivedcardiomyocytes evaluated by patch-clamp technique

H. Sadraei^{1,*}, S. Abtahi², M. Nasr Esfahani³, H. Baharvand³

¹Department of Pharmacology, School of Pharmacy, Isfahan University of Medical Sciences, Isfahan, Iran ²AJA University of Medical Sciences, Tehran, Iran ³Department of Cell and Molecular Biology, Royan Institute of Animal Biotechnology, Isfahan, Iran

Background and Aims: Development of embryonic stem cells to cardiomyocytes is accompanied with a great deal in ion channel expression and functions. In this research we have assessed K current development in Royan B1 cardiomyocytes using patch-clamp technique.

Methods: The mouse stem cells were differentiated into beating cardiomyocytes by hanging drop method. One week old stem cell derived cardiomyoctes (ESCs) were separated to single cell suspension for cell current recording. In this research we have investigated some of the properties of K current in ESCs using whole cell patch-clamp technique. The bath solution included 130 mM NaCl and 1.5 mM CaCl2. The intracellular pipette solution included 130 mM KCl, 3 mM ATP and 0.2 mM EGTA unless otherwise stated.

Results: At holding potential (HP) of -60 mV, in some cells a major outward current was elicited by square depolarizing pulses from -60 mv to +50 mV. This outward current was sustained for the duration of 300 ms test pulse. The sustained outward K current was inhibited by tetraethylammonium (TEA 10 mM) indicating the activity of Ca2+ activated K channel in these cells. Furthermore, when concentration of EGTA in the pipette was increased to 10 mM the sustained outward current was almost abolished, which again indicate that this K current is mainly elicited by Ca2+ ions. In some the cells with 0.2 mM EGTA in the cell we only get a very small outward current which suggest that in these cells the voltage activated K channels is absent or if exist it is not fully functional. Other cells were in far between indicating that voltage activated K channels are developing in these cells but it is not yet fully functional.

Conclusions: In conclusion, we have identified the expression of large conductance Ca2+ activated K channel in ESCs. However, our research shows that not all the stem cells are fully developed to cardiomyocyte.

Keywords: Embryonic stem cells; Royan B1 line; Cardiomyocytes; K⁺ current; Patch-clamp