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Postnatal developmental changes in the TRPC channels in mouse cardiomyocytes

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Introduction:
Evidence has been presented to indicate that there are postnatal developmental changes in cardiac Ca2+ channels and transporters, such as sarcolemmal L-type channels, Na+/Ca2+ exchangers, and the sarcoplasmic/endoplasmic reticulum Ca2+ ATPase (SERCA2a). The transient receptor potential canonical (TRPC) channels are Ca2+-permeable nonselective cation channels widely expressed in diverse cell types including cardiac myocytes and are typically activated following depletion of endoplasmic/sarcoplasmic reticulum (ER/SR) Ca2+ stores. The present study was undertaken to investigate the possible developmental changes in TRPC channels in mouse ventricles.
Methods:
Single ventricular myocytes were enzymatically isolated from various postnatal stages (day-0 to adult) of C57BL/6J mice using a retrograde Langendorff perfusion method. Membrane currents were measured using whole-cell patch-clamp methods with Cs+-rich pipettes and K+-free bath solution and were expressed as current density (pA/pF). The expression of TRPC isoforms was quantitatively assessed by Western blotting.
Results:
Bath application of thapsigargin (2 μM) gradually activated the membrane current that exhibited a practically linear I-V relationship with a reversal potential of ~0 mV, a sensitivity to inhibition by 2-aminoethoxydiphenyl borate (20 μM, 2-APB) and marked reduction in inward current after replacement of extracellular Na+ with N-methyl-D-glucamine (NMDG). Based on these properties, the thapsigargin-induced current was identified as the TRPC current. The amplitude of TRPC current measured at +40 mV peaked in day-0 neonatal ventricular myocytes (5.8±1.2 pA/pF) and declined thereafter during postnatal development to 1.8±0.9 pA/pF in adult ventricular myocytes. Western blot analysis revealed that expression of TRPC1, 3, 4, 5 proteins was also gradually decreased during development from neonate to adult.
Conclusions:
Our results provide experimental evidence to show that expression of TRPC channel is developmentally regulated in the mouse ventricle with a higher level in neonatal compared with adult ventricular myocytes.