fluorescence imaging. Experimental variables (spontaneous rate, AP amplitude and duration, CT decay time course) are extracted and compared to the output of the computational model. Conversely, results of the computational model (upregulation of If and ICa and downregulation of IK1 and Ito) into the model showed the presence of automaticity with the characteristic slow late diastolic depolarization. The maximum activation frequencies (0.7Hz) reached only the lower frequency range documented experimentally and found in the literature (0.4 to 6Hz).

Conclusion. Limitations in the rate of automaticity found emphasise the importance of exploring the Calcium Clock mechanism whose insertion may lead to higher spontaneous rates. In the future, a bifurcation analysis will give some insights on the dynamical mechanisms behind pacemaker generation in cultured NRVMs.

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Calcium Signaling Properties of Control and CPVT-Expressing Human iPSC-Derived Cardiomyocytes
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Recent advances in the field of stem cell biology have made it possible to develop spontaneously beating myocytes, with cardiac electrophysiological and molecular phenotypes, from skin fibroblasts. Little is reported, however, on Ca-signaling properties of such cells. Here we quantify the Ca-signaling parameters of human iPSC-derived cardiomyocytes developed from control and CPVT-affected subjects with F2483I point mutation in the RyR1 gene. Single 3-6 days hiPSCs were patch-clamped using pipette solutions containing in mM: 0.1 fluo4, 0.1 Ca2+, 0.2 EGTA, 5 Na+, 1 Ca2+ and global Ca-transients were simultaneously measured. Ca-buffer concentrations were set such as to maintain the spontaneous beating frequencies of intact cells. ICa averaged ~8 pA/pF in 3 hiPSCs cell-lines (2 controls & one CPVT-mutant), but with significant variation among cells (2-20 pA/pF). ICa-induced Ca-transients had rapid release but slow uptake kinetics, producing significant maintained components. The voltage-dependence of ICa-activated Ca-transients was bell-shaped, reflecting the voltage-dependence of ICa in both control and mutant cells. The ratio of ICa activated to caffeine-triggered Ca-transients (efficiency of release) was ~0.3. ICa induced by caffeine-transients ranged ~2 pA/pF compared to ~1.0 pA/pF in adult cardiomyocytes. The gain of CICR was voltage dependent, similar to adult cardiomyocytes. Both adrenergic and Ca-channel agonists enhanced ICa markedly, but failed to alter the gain of CICR significantly. Although there were no qualitative differences in Ca-signaling profiles of control and CPVT mutant myocytes, caffeine-triggered Ca-stores were smaller, CICR gain was larger at ~30 but not at 0mV, and Ca-sparks longer in duration in mutant myocytes.

We concluded that Ca-signaling parameters of hiPSCs-derived cardiomyocytes were similar to those of adult cardiomyocytes, but the CPVT-RyR mutant myocytes had somewhat altered Ca-signaling characteristics consistent with functionally defective RyRs. (Support by: NIH, ROI-HL16152, RO1-HL107600).

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Investigation of the Influence of hERG 1b on hERG Channel Pharmacology
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hERG channels mediating the rapid delayed rectifier K+ current (IKr) are important for normal ventricular repolarization. In native cardiac tissues, hERG 1a subunit co-assembles with a subunit encoded by an alternate transcript, “hERG1b” which has a shorter N-terminus (1) and influences hERG channel underlying IKr. hERG potassium channels contain nearly 300 different disease-causing mutations, which can lead to long QT syndrome (LQTS). Its channel deactivation properties. R56Q is a point mutation located in the PAS domain have different properties than those in the cell center; (2) in disease state, channels in gap junction coupling occur in tandem with alterations in Na+ channel density, Na+ channel properties, and subcellular structure, which makes it difficult to determine the effects of each change. To gain a greater quantitative understanding of the factors that influence CV, we implemented a one-dimensional mathematical model of electrically coupled human ventricular myocytes. The model accounts for the possibility that electrical fields in narrow clefts between adjacent cells can influence conduction. To determine how each model variable affects CV, we performed parameter sensitivity analysis under two conditions: (1) normal gap junctional conductance (400 kΩ resistance between cells), and (2) severely reduced gap junctional conductance (40 MΩ resistance between cells). Our simulation results predict the following: (1) Increasing the width of the cleft between adjacent cells increases CV under normal gap junctional conductances but decreases CV when gap junctional coupling is low; (2) reducing cell coupling decreases CV under normal gap junctional conductance conditions, but further changes to coupling have little effect under low gap junctional conductance conditions, (3) Na+ channel activation kinetics and voltage dependence have a greater quantitative effect on CV with low than with normal gap junctional conductance. The simulations generate predictions that can be tested experimentally, and the results provide insight into changes in CV observed in disease.  

EAG Domains Regulate LQT Mutant hERG Channels in Induced Pluripotent Stem Cell-Derived Cardiomyocytes
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From previous experimental and theoretical studies, it is well-known that gap junctional coupling, Na+ channel density, and Na+ channel localization influence the speed of electrical conduction velocity (CV) in multicellular cardiac tissue. However, two recent findings complicate our understanding of electrical conduction: (1) Na+ channels located at the intercalated disk region have different properties than those in the cell center; (2) in disease state, channels in gap junctional coupling occur in tandem with alterations in Na+ channel density, Na+ channel properties, and subcellular structure, which makes it difficult to determine the effects of each change. To gain a greater quantitative understanding of the factors that influence CV, we implemented a one-dimensional mathematical model of electrically coupled human ventricular myocytes. The model accounts for the possibility that electrical fields in narrow clefts between adjacent cells can influence conduction. To determine how each model variable affects CV, we performed parameter sensitivity analysis under two conditions: (1) normal gap junctional conductance (400 kΩ resistance between cells), and (2) severely reduced gap junctional conductance (40 MΩ resistance between cells). Our simulation results predict the following: (1) Increasing the width of the cleft between adjacent cells increases CV under normal gap junctional conductance conditions but decreases CV when gap junctional coupling is low; (2) reducing cell coupling decreases CV under normal gap junctional conductance conditions, but further changes to coupling have little effect under low gap junctional conductance conditions, (3) Na+ channel activation kinetics and voltage dependence have a greater quantitative effect on CV with low than with normal gap junctional conductance. The simulations generate predictions that can be tested experimentally, and the results provide insight into changes in CV observed in disease.

EAG Domains Regulate LQT Mutant hERG Channels in Induced Pluripotent Stem Cell-Derived Cardiomyocytes
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The generation of human induced pluripotent stem (hiPS) cells provides a novel path for a wide range of disease research, including LQTS. The human ether-a-go-go related gene (hERG) encodes the α-subunit of a voltage-gated potassium channel underlying IKr. hERG potassium channels contain nearly 300 different disease-causing mutations, which can lead to long QT syndrome (LQTS). Its N-terminal region contains an eag domain, which is important for modulating channel deactivation properties. R56Q is a point mutation located in the PAS domain that is associated with LQTS, a defect known to increase the rate of deactivation profoundly. Previous work showed that R56Q profoundly increased the rate of deactivation and lessened steady-state inactivation in Xenopus oocytes and mammalian HEK293 cells. We also showed that a soluble eag domain could restore the aberrant deactivation kinetics and inactivation properties of hERG R56Q. Here, we tested whether the delivery of adenosinergic eag domains could rescue hERG R56Q channels expressed in cardiomyocytes derived from human iPSCs (hiPSC-CMs). We used whole-cell patch-clamp recordings to measure currents from iPSC-CMs expressing hERG, hERG R56Q and hERG R56Q-eag domains. We found that the adenosinergic delivery of hERG R56Q to iPSC-CM resulted in channels with a faster rate of deactivation compared to that of WT hERG, indicating that hERG R56Q retained its deactivation. We also found that adenosinergic delivery of eag domains restored slow deactivation kinetics to hERG R56Q channels in iPSC-CMs. The results indicated that adenosinergic delivery of a small polypeptide (i.e. eag domain) could restore the current deficiencies in a hERG LQT-mutant channel. This result indicates that eag domain polypeptides may be a biological therapeutic for LQTS.