A Combined Gene and Cell Therapy Approach for Conduction System Repair

Anat Hofshi, Gil Arbel, Lior Gepstein

The Sohnis Laboratory for Cardiac Electrophysiology and Regenerative Medicine, and the Cardiology Department, Rambam Medical Center, The Bruce Rappaport Faculty of Medicine, Technion - Israel Institute of Technology, Haifa, Israel

Introduction: Impaired myocardial conduction may underlie both bradyarrhythmias and reentrant tachyarrhythmias. In the current study, we introduce a novel strategy for conduction system repair utilizing genetically engineered cells designed to form biological "conductive cables".

Methods and Results: An *in vitro* model of conduction block was established using spatially-separated, spontaneously contracting, non-synchronized, human embryonic stem cell-derived cardiomyocyte clusters. We next examined the hypothesis that HEK293 cells transfected with the $Na_v1.5$ voltage-gated sodium channel can couple with the cultured cardiomyocytes and synchronize the electrical activity of these spatially separated clusters. Cx43 immunostaining and Calcein-dye transfer experiments in co-culture studies confirmed formation of functional gap junctions between the engineered cells and neighboring cardiomyocytes. We next assessed the ability of the engineered cells to synchronize contractions between the separate clusters using a microelectrode array mapping (MEA) system. Synchronization was defined by the establishment of fixed local activation time differences (ΔLATs) between the two separate clusters and convergence of their spontaneous activation cycle lengths. Nontransfected control cells were able to induce synchronization between cardiomyocyte clusters separated by distances up to 200 μm. In contrast, the engineered cells synchronized contractions between cardiomyocyte clusters separated by up to 1000 μm, the longest distance studied. Finally, engineered cells expressing K^+ (Kv1.3) channels failed to induce any synchronization.

Conclusions: Genetically engineered cells, transfected to express Na^+ channels, can form biological conduits bridging and coupling excitable cells, allowing synchronization of contractions between distinct, widely separated cardiac cell clusters.