Activity of Self-Depolarizing Pacemaker Cells Bio-Engineered in Virtual Heterogeneous Ventricular Tissue

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Abstract

Genetically engineered cardiac pacemakers are a proposed alternative or support to the state-of-the-art electronic devices. Down-regulation of the expression of Kir2.1 channels reduces the activity of the inward rectifier current $\mathrm{I_{K1}}$ and turns ventricular cardiomyocytes to pacemaker cells.

This effect was mimicked in computational cells describing heterogeneous human ventricular electrophysiology by blocking the $\mathrm{I_{K1}}$ current. These intrinsic self-depolarizing cells were inserted into a long and thin parallelepipedal anatomical structure describing a ventricular strand from endocardium to epicardium. The width and position of the pacemaking region was variable.

The required width for successfully driving the tissue mainly depended on the amount of adjacent normal ventricular myocytes. Attachment of the pacemaker region to any tissue boundary reduced the necessary size. The smallest width was required for attachment to the endocardium. The cycle length of spontaneous activation was smallest for endocardial attachment and decreased with the size of the self-depolarizing region.

1. Introduction

Implantation of cardiac pacemakers is a common treatment for patients suffering from sick sinus node syndrome or conduction blocks. In the last years, strategies using genetically engineered ventricular instead of electronic pacemakers were proposed [1, 2]. The expression of the Kir2.1 channel was down-regulated leading to reduced maximum conductance of the time independent inward rectifying membrane current $\mathrm{I_{K1}}$. The affected cardiomyocytes turned from ventricular cells to pacemaking cells and developed spontaneous and rhythmic action potentials. Compared to electronic devices these cells would not require an elaborate surgical implantation, do not depend on a battery, may be regulated by the autonomous nervous system, and may adapt to changes of hormones.

We investigated the effects of Kir2.1 down-regulated computational cells within a virtual ventricular tissue including electrophysiological heterogeneities. The minimum size of the biologically engineered pacemaking area leading to successful driving of the surrounding tissue was investigated depending on its transmural position in a parallelepipedal strand model. Furthermore, the intrinsic pacemaking frequency of these cells was compared to the frequency in the driven coupled tissue.

2. Methods

The electrophysiological basis of this study is a computational model of a human ventricular myocyte [3]. This model describes ionic currents, ionic concentrations, and transmembrane voltage for one single virtual cell by a set of coupled nonlinear ordinary differential equations. These equations were solved in double precision by a forward Euler method with a time increment of 20 μs. The considered unequal ion channel characteristics of endocardial, epicardial and M cells were expanded by a transmural heterogeneous description of the maximum conductance $\mathrm{k_{NaCa}}$ of the sodium-calcium exchanger current [4] with the default conductance of the current describing epicardial electrophysiology. In contrast to a previous study modeling mutant inward rectifier $\mathrm{I_{K1}}$ channels [5], a model mimicking bio-engineered down-regulated $\mathrm{I_{K1}}$ channels was generated by reducing the maximum conductance $\mathrm{g_{K1}}$ of the $\mathrm{I_{K1}}$ current to 0% in all cell types. To achieve steady-state conditions this cellular model was computed once for 100 s and the state variables at the time of the last maximal diastolic potential were set as new initial conditions for the further simulations. The intrinsic cycle duration of the spontaneous depolarization was calculated based on the temporal difference of the peak values of the transmembrane voltage. Initial values for the states in the physiological non-blocked virtual cells were achieved by pacing the model with a frequency of 2 Hz for 100 s.

The single cell environment was transferred to a multicellular model of excitable tissue. Isotropic electrical coupling between these cells was obtained by using the mono-

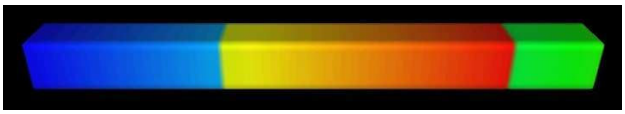

Figure 1. Exemplary tissue distribution in the transmural strand. Dark color shows normal, non-blocked tissue describing human cellular electrophysiology from endocardium (blue) to epicardium (green). Bright color visualizes the area of I_{K1}-blocked cells with intrinsic spontaneous pacemaking activity. The center and width of this area is variable.

domain reaction-diffusion model [6]. Its equations were solved in double precision by an iterative Gauss-Seidel method every 20 μs. Smooth transitions for the heterogeneously distributed ion channel characteristics were included in the underlying model of cellular electrophysiology. The virtual M cells were located near to the endocardium to obtain a realistic upright positive T wave in the transmurally computed ECG after endocardial stimulation [7].

A long and thin parallelepiped describing a ventricular strand from endocardium to epicardium formed the underlying anatomical structure. It consisted of 96x9x9 cubic volume elements (voxel) with an edge length of 0.2 mm. The resulting width of 19.2 mm corresponded to measurements. The conduction velocity of a planar wave in this tissue consisting of normal non-blocked cardiomyocytes was 0.62 m/s.

A range inside the normal ventricular tissue was replaced by self-depolarizing cells. The electrophysiological configuration of these cells corresponded to the configuration of the source cells except the I_{K1} block and the adjusted initial conditions. The center of this area as well as its spatial dimension in transmural direction was altered with a stepsize of one voxel (Fig. 1). The required size of the tissue depending on its transmural position to achieve a successful activation was calculated in different configurations to obtain a statistical analysis about the ability of the g_{K1} down-regulated cardiomyocytes to drive the surrounding heterogeneous tissue.

3. Results

Blocking of g_{K1} to 0% modified the characteristics of the ventricular cells. The stability of the resting potential by the inward rectifier I_{K1} was disabled and the resulting net inward current raised the transmembrane voltage of the virtual cells over time. The equilibrium potential of sodium and potassium affected currents was shifted due to drifts of the according ionic concentrations. The increased resting potential reduced the availability of the sodium channels. Thus, the cells required an increased

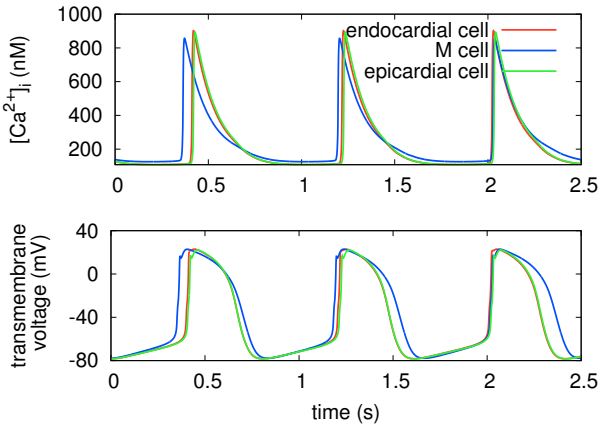

Figure 2. Time course of intracellular Ca^{2+}-concentration and transmembrane voltage in steady-state conditions during spontaneous depolarization. The Ca^{2+}-range remains constant leading to a stable pacemaker activity in the distinctive g_{K1}-blocked configurations.

stimulus current for the I_{Na} channel activation compared to normal cells. As soon as the threshold was reached a spontaneous depolarization of the cellular membrane occured. The model generated a stable range of the intracellular calcium concentration and a non-self-terminating pacemaker activity in all cell types (Fig. 2).

The electrophysiological heterogeneities in the distinctive cell types led to different characteristics in the g_{K1}-blocked configuration. As in the non-blocked situation the action potential duration was longest in the M cells and epicardial as well as M cells developed a more pronounced notch in the early phase of the repolarization (Fig. 2). Although the maximal diastolic potential showed barely variations in transmural direction, the cycle length of the spontaneous activation was 794 ms in virtual M cells and decreased to 757 ms and 740 ms in endocardial and epicardial direction, respectively (Fig. 3).

In the multi-cellular tissue model a large amount of g_{K1}-blocked cardiomyocytes was required for successful activation of the adjacent normal tissue. The surrounding tissue pulled down the resting potential of the pacemaking cells due to electrotonic coupling if the blocked sector was too small. The net inward current during diastole was not large enough to raise this transmembrane voltage over the threshold value for I_{Na} activation. Spontaneous activity in these intrinsic self-depolarizing cells was therefore inhibited. The electrotonic interactions were reduced and spontaneous activity was no more suppressed if the blocked area exceeded a critical amount (Fig. 4).

We computed several distributions of the pacemaking cells within the transmural ventricular strand (Fig. 1). The required size of the area for successful pacing was smaller

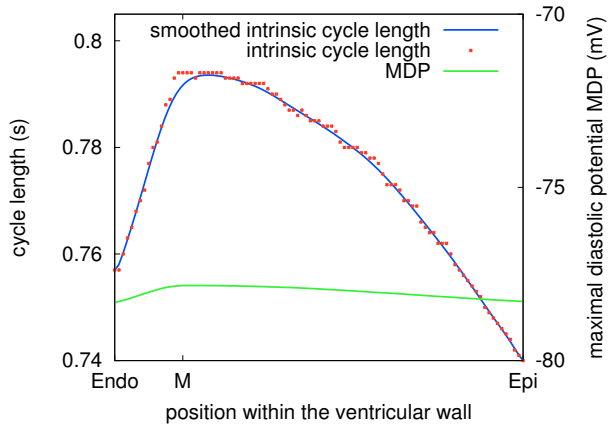

Figure 3. Intrinsic cycle length and maximal diastolic potential of g_{K1} down-regulated cardiomyocytes in the single cell environment. The cycle length was largest in the area of the M cells. The highest frequency was achieved within g_{K1}-blocked epicardial cells.

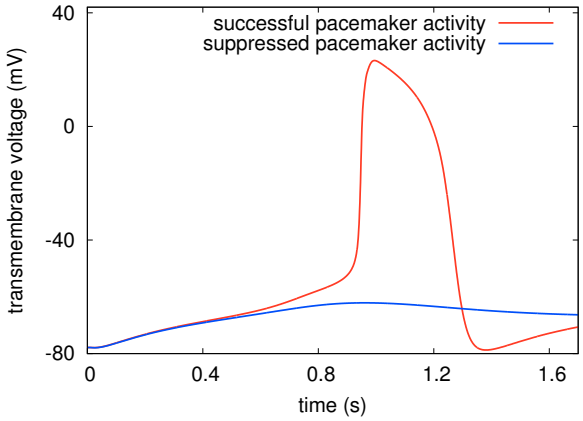

Figure 4. Temporal variation of the transmembrane voltage within the g_{K1}-blocked area in the multi-cellular environment. The influence of the increased inward current was reduced and spontaneous activity was suppressed if the area of the pacemaking tissue was too small. In a larger amount of pacemaking tissue the threshold value for I_{Na} activation was reached and the depolarization spread in the adjacent ventricular tissue.

if it was attached to the endocardial or epicardial boundary compared to a region of self-depolarizing cells completely embedded in normal tissue. Attachment of the area to the endocardium required a minimum size of 11.2 mm for successful stimulation of surrounding tissue. This critical size was slightly increased to 11.8 mm if the pacemaking region was attached to the epicardial boundary. The activation started nearby the considered tissue boundary and nearly simultaneously in the area of blocked and non-blocked interconnection (Fig. 5). The minimum size of the g_{K1}-blocked tissue was 18.4 mm if it had no contact to any outside surface. In this case, the depolarizing wave front startet not from inside the pacemaker but nearby the junction of the normal and pacemaking tissue (Fig. 5).

The resulting pacing frequency of the bio-engineered pacemaker was reduced in the multi-cellular tissue compared to the intrinsic frequency of the single g_{K1}-blocked cardiomyocytes. A pacemaking area attached to the endocardium led to a reduced cycle length compared to an area of self-depolarizing cells with the same size attached to the epicardium. The cycle period generated by a 12 mm wide pacemaking area was 1114 ms for endocardial and 1324 ms for epicardial attachment and was reduced with increased width of this area (Fig. 6).

4. Discussion and conclusions

Consistent with measurements [1, 2], suppressed activity of the inward rectifier current I_{K1} generated a net inward current during diastole and turned physiological ventricular cardiomyocytes to self-depolarizing pacemaker cells. While the maximal diastolic potential in the distinctive cell types showed only little variations, the net inward current and therefore the frequency of the spontaneous depolarization was determined by the position of the regarding cell within the ventricular wall (Fig. 3). Both were largest on the epicardial and endocardial tissue boundaries and dimished in midmyocardial direction. In contrast to a previous study using the Priebe-Beuckelmann model for describing cellular electrophysiology [8], we observed a stable non-self-terminating pacemaker activity.

The required width of the g_{K1}-blocked area within a heterogeneous ventricular tissue for successful driving this tissue as well as the resulting frequency of spontaneous activation were mainly determined by electrotonic interactions between ventricular and pacemaking tissue. The number of adjacent cardiomyocytes and therefore the electrotonic influence was reduced if the pacemaking area was attached to a tissue boundary. Thus, the required width of the pacemaking region was reduced in this case compared to the situation where the region was completely embedded in normal tissue. Furthermore, increased width raised the spontaneous frequency but reduced the differences between endocardial and epicardial attached configuration (Fig. 6). This is due to the fact, that in both cases these configurations tend to a fully g_{K1}-blocked tissue. The depolarizing wave front started either within the pacemaking tissue or nearby the interconnection of blocked and non-blocked cardiomyocytes. In the latter case, the lower threshold for I_{Na} activation in the adjacent ventricular tissue was reached due to electrotonic interactions of the pacemaker to the normal cells.

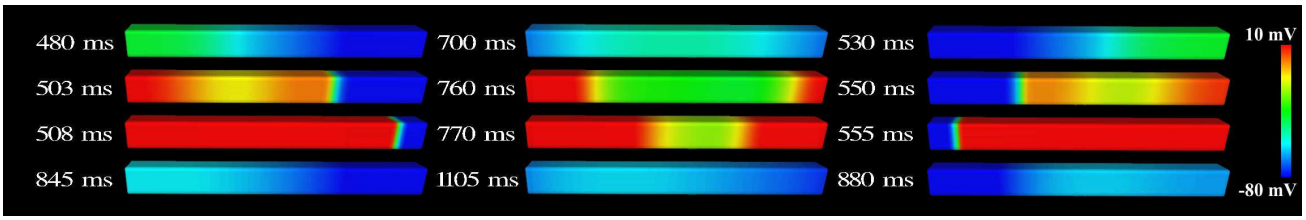

Figure 5. Transmembrane voltage distribution in the parallelepipedal anatomical structure including heterogeneous electrophysiology from subendocardium (left side) to subepicardium (right side). Bright color corresponds to activated tissue and dark depicts the resting potential of the cardiomyocytes. A 12 mm wide pacemaking region was attached to endocardium (left column) and epicardium (right column). The depolarization started nearly simultaneously nearby the corresponding tissue boundary and at the interconnection of blocked and non-blocked cells. In the center column, the 18.4 mm wide region consisting of engineered pacemaking cells had no contact to any outside surface. The depolarization started not from inside the pacemaker but nearby the junction of pacemaking and normal tissue.

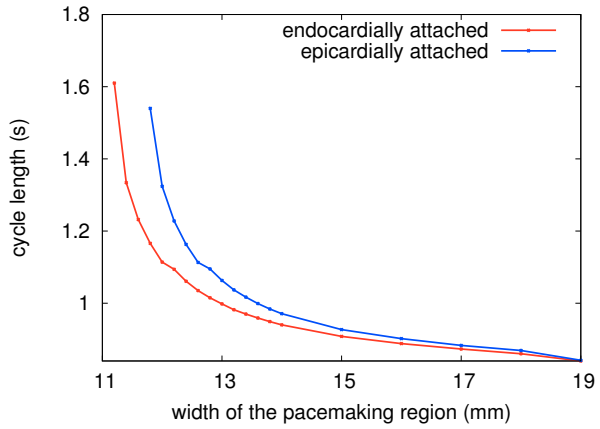

Figure 6. Cycle length in multi-cellular tissue. Endocardially g_{K1}-blocked cells developed a reduced cycle length compared to epicardially g_{K1}-blocked myocytes and required a less critical width of the pacemaking area to drive the surrounding tissue.

We determined the effects of g_{K1}-blocking in a heterogeneous cellular model describing the electrophysiology of the human ventricle. The resultant biologically engineered ventricular pacemaker would be an alternative or support to electronic devices in the case of excitation conduction blocks. As therapy for excitation initiation diseases, an atrial pacemaker would be the more consequential strategy. We will investigate the effects of I_{K1} reduction in atrial tissue in a future study. Spontaneous activity in the atrium due to the engineered pacemaker would conduct the electrical excitation via the AV node and the Tawara brunches leading to a physiological activation of the ventricles. Furthermore, we intend to investigate the influence of the extracellular conductance and the varying fiber orientation by using the bidomain approach in 2D as well as 3D anatomical structures.

References

[1] Miake J, Marban E, Nuss HB. Gene transfer: biological pacemaker created by gene transfer. Nature 2002;419:132–133.

[2] Miake J, Marban E, Nuss HB. Functional role of inward rectifier current in heart probed by Kir2.1 overexpression and dominant-negative suppression. J Clin Invest May 2003; 111(10):1529–1536.

[3] ten Tusscher KHWJ, Noble D, Noble PJ, Panfilov AV. A model for human ventricular tissue. Am J Physiol 2004; 286:H1573–H1589.

[4] Zygmunt AC, Goodrow RJ, Antzelevitch C. I_{NaCa} contributes to electrical heterogeneity within the canine ventricle. Am J Physiol 2000;278:1671–1678.

[5] Seemann G, Sachse FB, Weiss DL, Dössel O, Tristani-Firouzi M. Arrhythmogenic effects of cardiac IK1 mutations in virtual left ventricular tissue. Paper in preparation.

[6] Henriquez CS, Muzikant AL, Smoak CK. Anisotropy, fiber curvature and bath loading effects on activation in thin and thick cardiac tissue preparations: Simulations in a three-dimensional bidomain model. J Cardiovasc Electrophysiol May 1996;7(5):424–444.

[7] Weiß DL, Seemann G, Dössel O. Conditions for equal polarity of R and T wave in heterogeneous human ventricular tissue. In Proc. BMT, volume 49-2/1. 2004; 364–365.

[8] Zhang H, Tong WC, Garratt CJ, Holden AV. Stability of genetically engineered cardiac pacemaker – role of intracellular Ca^{2+} handling. In Proc. Computers in Cardiology, volume 32. 2005; 969–972.

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