Effect of percentage reduction in Action Potential Duration of M cells on reentry in short QT syndrome

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Abstract

Increase in transmural dispersion of repolarisation along with a diminished QT interval have been known to aid in the development of arrhythmia during KCNQ1-linked short QT syndrome type 2 (SQTS2). However, the percentage by which action potential duration (APD) shortens in the different cell types that make up the ventricular wall are not fully understood. In this study, the percentage of APD shortening of M cells was varied to determine the conditions under which re-entry occurs during SQTS2. A 2D transmural section of the heart with anisotropic properties is considered. Slight modifications to the TP06 equations are used to simulate the electrophysiology of the endocardial (endo), midmyocardial (M) and epicardial (epi) cells. A discrete network of 250×100 cells are interconnected using gap junction conductances and from this, a pseudo ECG is generated. On pacing the tissue with premature beats in the midst of normal pacing pulses and on including SQTS, it is observed that re-entry is sustained for a longer duration when the APD shortening in M cells is more compared to the epi or endo cells while the percentage reduction in APD of M cells is about 5% to 7% lesser than that in epi and endo cells. Further, when the percentage reduction in APD of M cells is similar to epi or endo cells, no re-entry is generated. This analysis highlights the key role of percentage reduction in APD of M-cells compared to epi and endo cells in maintaining the re-entrant waves.

1. Introduction

Appearance of a abnormally short QT interval in combination with tall peaked T-waves on the electrogram is classified as Short QT syndrome (SQTS). The shortened refractory period of cells during SQTS causes an increased vulnerability to ventricular arrhythmias. Six type of genetic variants of SQTS have been identified up to date with mutations in the genes encoding the different potassium currents or L-type calcium current [1]. In this paper, the focus is on SQTS2 caused due to a mutation in KCNQ1, which encodes the α-subunit of the slow delayed rectifying potassium current (I_{Ks}). Zhang et al. [2] developed Hodgkin-Huxley type equations to address the different types of mutations in I_{Ks} channel and the same is adapted here.

KCNQ1 mutation increases the risk to arrhythmogenesis due to an increase in transmural dispersion of repolarisation (TDR) in the tissue. The difference in repolarisation times of the different types of cells: endocardial (endo), midmyocardial (mid) and epicardial (epi) layers that make up the ventricular wall exist due to the inherent difference in ionic properties. In addition to these three layers, a major influencer of TDR is the presence of M-cells along the deep subendocardium to midmyocardium [3]. On slowing of heart rate or on exposure to QT prolonging drugs like sotalol, quinidine etc., these M-cells substantially prolong their action potential duration (APD) compared to endo or epi cells thereby creating a substrate for arrhythmogenesis. The ionic mechanisms underlying the different variants of SQTS and as well as its proarrhythmic effect in 2D and 3D realistic anatomical geometries have been critically analysed in multi-scale tissue models [1, 2]. However, the above studies do not critically analyse the properties of M-cells in relation to the surrounding epi and endo cells in generating an arrhythmia. In this paper, a transmural anisotropic tissue grid is considered incorporating the different types of cells. The APD of the M-cells alone is varied under SQTS2 conditions to analyse the percentage of M-cell APD shortening that can trigger a ventricular arrhythmia.

2. Methods

The biophysical ventricular cell model developed by Ten Tusscher et al. [4] is used to describe the electrical activity of the endo, mid and epi cells. Modifications to this model were made as done in the study by Priya et al. [5] so that the cells can develop early after depolarisations (EADs). The stimulus current has an amplitude of 52 μA and is applied for 1 ms. The differential equations of the ionic variables are integrated with a time step of 0.05 ms. An array of cells having a length of 250 cells and a thick-

ness of 100 cells is used to represent a section of a 2D transmural ventricular wall as was developed by Priya et al. [5]. Cells are interconnected by conductances that mimic the function of gap junction. The origin (Cell 1,1) in this array of cells is taken as the bottom leftmost corner. Starting from the left side of the tissue, the first 25% column of cells are considered as endo cells, the next 35% is mid cells and the the remaining 40% is epi cells. Anisotropy is included in the tissue by setting the gap junction conductance (GJC) along the length and thickness of the tissue to 4 μS and 0.4 μS so that the conduction velocity along the length and thickness is 76.9 cm/s and 33.3 cm/s respectively. The GJC at the mid-epi interface is reduced by 5 times so as to mimic those observed experimentally by Gima et al. [6]. There is no reduction in GJC at the endo-mid interface. The entire mid layer is considered to be made up of M-cells which have longer APD compared to endo or epi cells. A small group of cells (Cells 1:10,1:2) in the lower leftmost corner of the tissue is considered as the pacing site and is excited every 800 ms. Normalised Pseudo ECGs are then created based on the equations described by Gima et al. [6].

The mutation induced changes in I_{Ks} current are incorporated by changing the parameters of equations for I_{Ks} based on the experimental data on SQT2 KCNQ1 V307L obtained by Bellocq et al. [7]. Four different KCNQ1 V307L mutation induced changes are considered: wild type (WT), heterozygous (Het), homozygous (Hom) and homozygous reduced (HomRed). The equations of I_{Ks} current are described by Eqns. 1-3. The time constant of x_s-gate of I_{Ks} current in Eqn. 4 is multiplied by a term 'Fac' whose values are summarised in Table 1 for the different cell types and gene mutations. The equations for steady state of x_s-gate for the different gene mutations is given in Eqns. 5-7. Additionally, the I_{Ks} current in epicardial cells is scaled by a factor of 1.5 compared to midmyocardial cells as reported in the study of Szabo et al. [10], so as to generate a high, symmetrical T-wave as observed in clinical ECG under SQTS conditions.

$$I_{Ks} = G_{Ks} x_s^2 (V_m - E_K) \tag{1}$$

$$\alpha_{xs} = \frac{1400}{\sqrt{(1 + e^{(5 - V_m)/6})}} \tag{2}$$

$$\beta_{xs} = \frac{1}{(1 + e^{(V_m - 35)/15})} \tag{3}$$

$$\tau_{xs} = Fac * (\alpha_{xs}\beta_{xs} + 80) \tag{4}$$

$$WT : x_{s\infty} = \frac{1}{(1 + e^{(-5.9 - V_m)/17.4})} \tag{5}$$

$$Het : x_{s\infty} = \frac{1}{(1 + e^{(-20.62 - V_m)/10.96})} \tag{6}$$

$$Hom : x_{s\infty} = \frac{1}{(1 + e^{(-24.05 - V_m)/16.09})} \tag{7}$$

$$HomRed : x_{s\infty} = \frac{1}{(1 + e^{(-24.05 - V_m)/16.09})} \tag{8}$$

Table 1: 'Fac' value used for different mutations

	Fac for M cells	Fac for Endo/Epi cells
WT	1	1
Hetero	0.75	0.7
Homo	0.58	0.52
HomRed	0.38	0.32

In order to study the percentage of APD shortening of M-cells that triggers an arrhythmia, heterozygous SQTS2 mutation is introduced in all the cells of the tissue. The term 'fac' used in Eqn. 4 to calculate the time constant of x_s gate of I_{Ks} current is varied only for M-cells to different values: 0.6, 0.65, 0.7, 0.75 and 0.8 so that the APD of the M-cell changes. Further, in between the normal pacing pulses, premature beats are introduced in between in order to test if an arrhythmia can be generated under het SQTS2 mutation.

3. Results and Discussion

The different SQTS2 gene mutations are introduced one at a time in all the cells of the tissue and then the tissue is stimulated at the normal pacing site at a heart rate of 75 beats/min beats. A convex propagating depolarisation pattern arises moving from the endo to the M and epi layers from the bottom to the top of the tissue. Repolarisation occurs in the epi and endo cells and the M-cells are the last to repolarise. The pseudo ECGs for the different gene mutations are plotted in Fig. 1. The difference in repolarisation times between cells creates a positive T-wave. The QT interval (QT_i), T-peak and TDR values are listed in Table 2. TDR is calculated as the time difference between the T_{peak} to T_{end}. It is observed that as the gene mutation changes from WT to Het, Hom and HomRed, the QT interval reduces while the T-peak and TDR values increase.

Table 2: Parameters of different SQTS2 gene mutations

Gene Mutation	QT_i (ms)	T_{peak} (mV)	TDR (ms)
WT	350	0.4038	50
Het	300	0.435	55
Hom	275	0.4716	60
HomRed	245	0.485	65

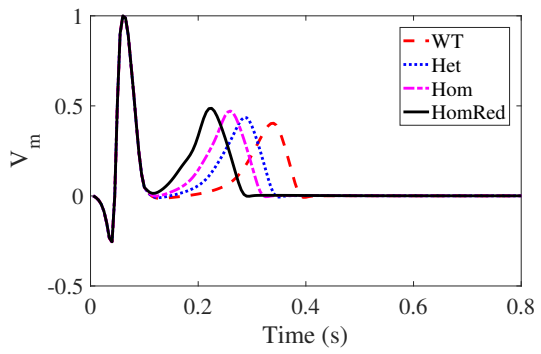

Figure 1: Pseudo ECG for different gene mutations

3.1. APD in single Cells

The SQTS2 Het gene mutation is incorporated in the three cell types and the APD of the cells is calculated. The 'fac' for endo and epi cells is a constant value of 0.7 while that of M-cells is considered for five different values: 0.6, 0.65, 0.7, 0.75 and 0.8. The difference in APD between WT and Het gene mutation and the corresponding percentage change in APD (PC_{APD}) is given in Table. 2. The PC_{APD} in M-cells is almost similar to that in endo and epi cells when the 'fac' is 0.6. When the 'fac' is increased, the APD of the M-cells increases while the corresponding difference in APD and percentage change in APD are shown to decrease.

Table 3: Comparison of APD of different cell types between WT and Het gene mutation

	APD in WT (ms)	APD in Het (ms)	fac	△APD (ms)	PC_{APD}
Endo	281.8	225.4	0.7	56.4	20
Epi	251.9	198.2	0.7	53.7	21.3
M	362.6	288.8	0.6	73.8	20.35
		296.6	0.65	66	18.20
		303.9	0.7	58.7	16.18
		310.7	0.75	51.9	14.31
		317.1	0.8	45.5	12.55

3.2. Generation of Arrhythmia

On including the SQTS2 Het gene mutation, the tissue is paced regularly every 800 ms. After six normal pacing pulses, three premature beats (PBs) each of 270 ms are applied inorder to test if an arrhythmia can be generated. Fig.2(i-iii) shows the normalised pseudo ECGs generated on application of this pacing protocol and on varying the 'fac' in M-cells from 0.6 to 0.65, 0.7, 0.75 and 0.8 respectively. It is observed that the first six beats in the pseudo ECGs are normal and appear at regular intervals of 800 ms. The percentage change in APD in M-cell is almost same to that of endo or epi cell when the 'fac' is 0.6. Here, the application of three PBs causes a positive QRS complex and an inverted T-wave as observed in Fig. 2(i) and the normal pacing pulse resumes at 5.6 s. The voltage snapshots of the tissue are shown in Fig. 2(iv)(a) after the application of the first PB at 4.27 s. At this time, the M-cells are still in repolarising state. Thus, the depolarisation proceeds upwards along the endo layer and then enters mid and epi layer as seen in Fig. 2(iv)(b). The cells located in the top of the mid and epi layer are the last to repolarise. A similar pattern of depolarisation and repolarisation are observed after applying the second and third PB at 4.54 s and 4.81 s respectively as seen in Figs. 2(iv)(c-h). This change in the repolarisation pattern causes an inverted T-wave complex to be generated in the pseudo ECG. Thus, no reentry is generated. When the 'fac' is 0.7, application of a single PB causes a upward-downward complex in the pseudo ECG as seen in Fig. 2(ii). When the third PB is applied at 4.81 s, a QRS complex with positive T-wave but a smaller amplitude is observed. Normal QRS complexes follow after 5.6 s. The voltage snapshots of the tissue are shown in Fig. 2(v)(a) after the application of the first PB at 4.285 s. As the mid cells are still repolarising, the wavefront arising from the PB travels upward along the endo layer and then moves right into the mid and epi layer at the top of the tissue as observed in Fig. 2(v)(b). This wave travels downward along the epi layer and reenters into mid and endo layer as seen in Fig. 2(v)(c). This reentrant wave continues in Fig. 2(v)(d-g) with all the cells returning to rest state just before the application of the third PB at 4.81 s in Fig. 2(v)(h). The third PB creates a normal depolarisation and repolarisation pattern and the normal rhythm is resumed at 5.6 s. Fig. 2(iii) shows the pseudo ECG on applying the same pacing protocol and when the 'fac' is increased to 0.75. On applying three PBs, an arrhythmic type of pattern is observed that is not sustained. The normal pacing pulse is resumed at 7.2 s after a long pause. Here, the percentage reduction in APD of M cells is about 5% to 7% lesser than that of endo or epi cells. Similar to the when 'fac' is 0.7, on applying a single PB, reentrant waves are generated when the M-cell 'fac' is 0.8. These waves continues when the second and third PB are applied and finally all cells repolarise at 5.37 s and the regular pacing pulse at 5.6 s creates a normal QRS complex (not shown). Increasing the 'fac' to 0.65 and on applying the same pacing protocol, reentrant waves are created after two PB and the this continues when the third PB is applied. However, all the cells get repolarised at 5.31 s and the normal pacing pulse is resumed at 5.6 s (not shown).

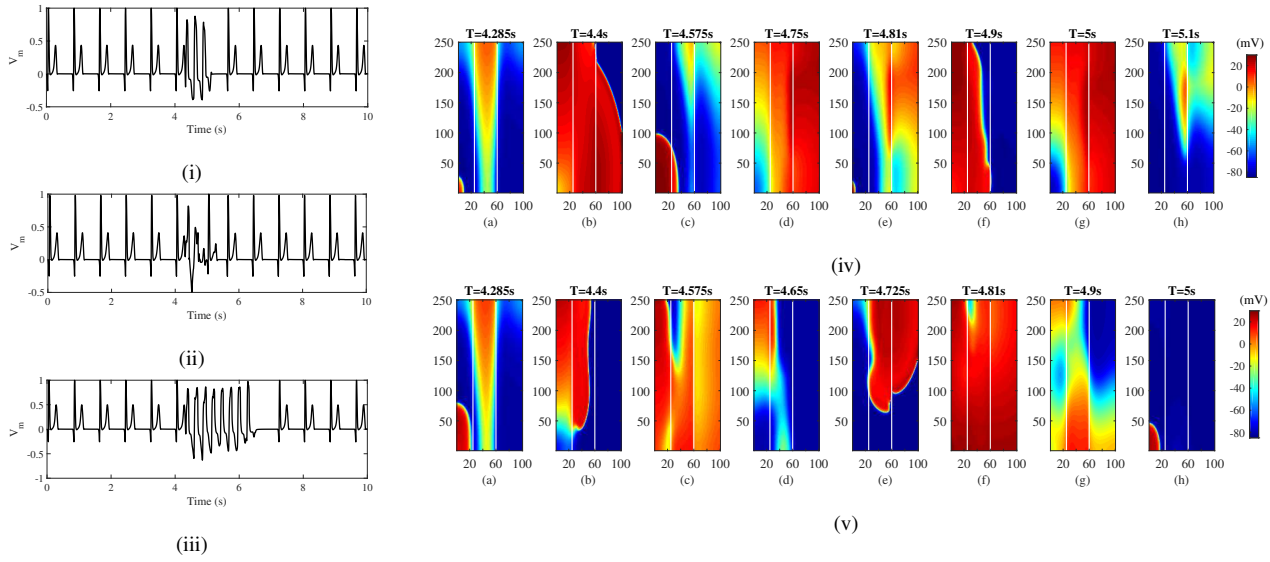

Figure 2: Pseudo ECG generated when M-cell 'fac' is (i) 0.6 (ii) 0.7 and (iii) 0.75 and Voltage snapshots on application of premature beats when the M-cell 'fac' is (iv) 0.6 and (v) 0.7.

4. Discussion and Conclusions

Adeniran et al. [1] used a markov chain model to incorporate mutant I_{Ks} kinetics in action potential model to represent SQTS2 conditions. They had tabulated the APD of single cells in WT and Het mutation conditions as well as the change in the APD. However, percentage change in APD from WT to Het was not considered. On calculating this value, it was found to be 28.31 %, 21.65% and 28.59 % in epi, mid and endo cells. Thus, it supports the findings observed here that the % variation in M-cells is about 5% to 7% lesser than that of epi or endo cells and this scenario further supported reentrant wave formation in 2D and 3D models. In this study, it is noticed that when the % reduction in APD from WT to Het mutation is between this range of 5% to 7%, reentry is sustained for a longer time compared to when it is lesser or greater than this range. Further, when the percentage reduction in APD of M cells is similar to that in endo or epi cells, no reentrant waves are generated. Therefore, this study highlights how the variation in APD shortening between M-cells and its surrounding endo and epi cells can act as a potential substrate for generating arrhythmia. A limitation of this study is that a simplified geometry is considered with the entire mid layer made up of M-cells while the M-cells have been reported to be present as islands. Further, the percentage variation in APD between M cells and other cell types have been compared only between WT and Het gene mutation. Other gene mutation: Hom and HomRed will also need to be analysed.

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