In Silico Optogenetic Control of Spiral Waves in GtACR1-transduced Atrial Diffuse and Focal Fibrotic Tissues

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Abstract

Atrial fibrosis is a significant factor in inducing atrial fibrillation (AF). In recent years, optogenetics has been experimentally confirmed to effectively terminate AF in rodents, whereas its effectiveness in human AF cardioversion has not yet been clearly established due to considerations of experimental safety and technological challenges. As an important adjunct and complement to clinical experimental research, computational modeling can provide both phenomenological simulation and mechanistic explanation for the occurrence and progression of AF. This study employed GtACR1-transduced human atrial cells and two-dimensional models of human atrium with focal fibrosis and diffuse fibrosis in chronic AF, to investigate the effects of sub-threshold illumination on cellular electrophysiology and spiral wave dynamics. Our results showed that with the same proportion of atrial fibrosis and gap-junctional conductance, the transmembrane potential of atrial myocytes was compelled to align with the reversal potential of GtACR1 in sub-threshold illumination. This "optogenetic voltage forcing" phenomenon effectively thwarted the re-excitation of atrial myocytes. As a result, spiral waves were terminated in both pure atrial muscle and tissues exhibiting diffuse and focal atrial fibrosis.

1. Introduction

The global prevalence of atrial fibrillation (AF) has experienced a notable increase over the past three decades [1]. AF's onset and progression are intimately linked to electrical and structural remodeling of the atrium. Among these factors, fibrosis creates a substrate for AF initiation and perpetuation, while persistent AF further remodels atrial architecture [2]. In comparison to traditional electrical stimulation, optogenetics boasts superior spatial and temporal resolution, sensitivity, and efficacy. Research into optogenetically modulating cardiac electrophysiological properties has flourished in recent years, and how to apply this technology to terminate arrhythmias such as AF has also attracted widespread academic interest [3]. When compared to the widely used channelrhodopsin-2 (ChR2), Guillardia theta anion channelrhodopsin-1 (GtACR1) exhibits a more negative reversal potential and has the advantage of bidirectionally modulating action potentials (APs) [4]. Although optogenetics has demonstrated promising results in cardiac defibrillation experiments in rodents, concerns over potential immune reactions in humans have precluded its direct clinical application for human cardiac defibrillation [5]. As a vital complement and validation tool for clinical trials, computational modelling can further explore the parametric indicators necessary for achieving effective and safe optogenetic defibrillation in human hearts.

This study utilizes electrophysiological models of GtACR1-transduced human atrial myocytes and fibroblasts (Fbs), alongside two-dimensional (2D) atrial models featuring diffuse and focal fibrosis, to preliminarily investigate the effects of sub-threshold illumination on cellular electrophysiology and spiral wave dynamics under chronic AF conditions.

2. Materials and methods

Computational simulations were conducted via a combined model of GtACR1-transduced human atrial myocyte under chronic AF conditions [4, 6, 7]. For fibrotic regions, the model described above was coupled to an electrophysiological model of Fbs [8]. An overview of numerical study was given as follows.

2.1. Modeling of electrical activity in single atrial cells

The human atrial AP model under chronic AF conditions has been described in prior work [6, 7]. The

atrial Fb model contains 4 active membrane conductances, I_{Kv}, I_{K1}, I_{NaK} and I_{bNa} [8]. To include light sensitivity, the AF model is coupled to the mathematical model of the GtACR1 photocycles [4]. The time dependent change in single atrial myocyte's transmembrane potentials of atrial myocyte is described by Equation 1. Here, V_m represents the atrial myocyte membrane potential, C_m is the atrial myocyte membrane capacitance, I_m is the sum of all transmembrane ionic currents, I_{stim} is the externally applied electrical stimulus current, I_{GtACR1} is the current density of the channel GtACR1.

$$\frac{dV_m}{dt} = -\frac{1}{C_m}(I_m + I_{stim} + I_{GtACR1}) \quad (1)$$

If the atrial myocyte is assumed to couple with Fbs, electrical activities in atrial myocyte and Fb are modelled according to Equations 2 and 3. Here, V_{fi} represents the transmembrane potential across the ith coupled Fb, I_{fi} represents the current output for the ith coupled Fb, C_f is the Fb membrane capacitance, G_{gap} is an intercellular conductance, n is the total number of Fbs.

$$\frac{dV_m}{dt} = -\frac{1}{C_m}\left(I_m + I_{stim} + I_{GtACR1} + \sum_{i=1}^{n} G_{gap}(V_m - V_{fi})\right) \quad (2)$$

$$\frac{dV_{fi}}{dt} = -\frac{1}{C_f}\left(I_{fi} + G_{gap}(V_{fi} - V_m)\right) \quad (3)$$

2.2. Modeling of electrical activity in a 2D domain of the human atrium

We assumed three situations in 2D monodomain atrial modelling: non-fibrotic tissue, diffuse fibrotic tissue and focal fibrotic tissue. The domain consisted of 101 × 101 grid points with a spatial resolution of 0.025 cm. As shown in Figure 1, each point represented a single atrial cell or an atrial cell coupled with Fbs. The proportion of the two types of fibrosis in the atrium was set to 30% for both. For the diffuse fibrosis, 30% of the grid points (101 × 101 × 30%, nearly 3060 points) were randomly and uniformly selected across the entire area to represent atrial myocytes coupled with Fbs. For the focal fibrosis, 30% of the grid points were concentrated and selected within the centre of the region (approximately a square-shaped area) to serve as coupling points.

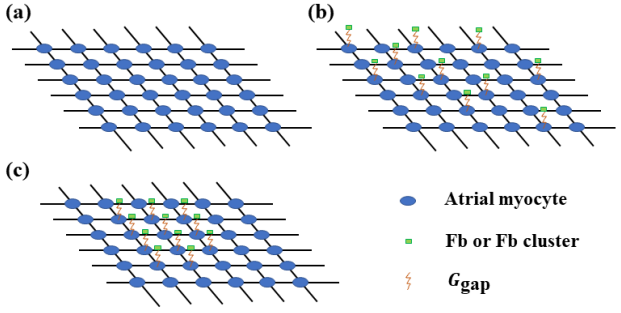

Figure 1. Schematic diagram of a small part of 2D simulation domain with sites occupied by (a) pure atrial myocytes, (b) atrial diffuse fibrosis and (c) atrial focal fibrosis.

The membrane voltage in spatially extended media described above was modelled via a reaction-diffusion type equation. Combining equations 1 and 2, the equation was written as follows. Here, D was the diffusion tensor and was assumed to be a scalar (0.00014 cm^2/ms). For fibrotic regions, conductivity values between a pure myocyte and a myocyte-Fb coupling and between two myocyte-Fb couplings were reduced by 10% and 30%, respectively.

Non-fibrotic region:

$$\frac{dV_m}{dt} = \nabla(D\nabla V) - \frac{1}{C_m}(I_m + I_{stim} + I_{GtACR1}) \quad (4)$$

Fibrotic region:

$$\frac{dV_m}{dt} = \nabla(D\nabla V) - \frac{1}{C_m}\left(I_m + I_{stim} + I_{GtACR1} + \sum_{i=1}^{n} G_{gap}(V_m - V_{fi})\right) \quad (5)$$

2.3. Simulation protocol

This study focused on the impact of photocurrent rather than fibrosis on spiral wave dynamics. Therefore, in the simulations of two atrial fibrosis models, G_{gap} and the number of Fbs (n) coupled to each atrial myocyte were set to fixed values (G_{gap} = 2.0 nS, n = 4).

The simulation program was written in Fortran language. At the cellular level, the forward Euler method was employed to update all state variables with the temporal resolution of 10^{-3} ms. Additionally, the finite difference method (FDM) was utilized to tackle the reaction-diffusion equation.

3. Results

3.1. GtACR1-based atrial APs in fibrotic and non-fibrotic regions

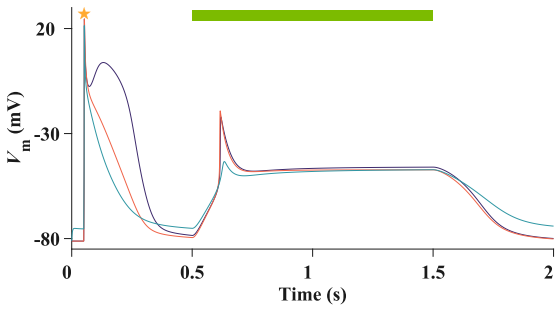

Figure 2. Atrial APs obtained in healthy tissue (purple), as well as non-fibrotic (red) and fibrotic tissue (blue) with AF. Timing of electrical stimuli and the interval of illumination (green light, 515 nm, 1000 ms, 0.001 mW/mm^2) were represented by an orange star and a green bar, respectively.

Figure 2 showed myocyte membrane kinetics in healthy tissue (purple), as well as non-fibrotic (red) and fibrotic regions (blue) under chronic AF conditions. After the electrical stimulation, the repolarization of pure atrial myocytes under chronic AF conditions and that with AF accompanied by fibrosis was significantly accelerated as compared to healthy atrial myocytes. Compared to non-fibrotic areas, the changes in AP of atrial myocytes in fibrotic areas were as follows: APD_{80} (AP duration at 80% repolarization) was shortened by 21.0%, and resting transmembrane potential as well as peak transmembrane potential were reduced by 7.4% and 12.7%, respectively. During the illumination, atrial myocytes in fibrotic areas exhibited the slowest upstroke velocity and repolarization, as well as the lowest peak potential.

3.2. Influence of sub-threshold illumination on spiral waves

Figure 3 depicted the dynamic changes of spiral waves under sub-threshold illumination in pure atrial myocardium, atrial diffuse fibrosis, and atrial focal fibrosis under chronic AF condition. Atrial myocytes in all three types of atrial tissues expressed the I_{GtACR1}. The spiral waves were initiated by S1-S2 electrical stimulation (first column) and then persisted in rotating in both pure atrial myocardium and diffuse fibrosis tissue (second column, Figure 3a-3b), but fragmented in focal fibrosis tissue (second column, Figure 3c). Upon the application of uniform global illumination at 0.001 mW/mm², the spiral waves in all three types of atrial tissues began to gradually dissipate (third column of the figure), ultimately disappearing (fourth column). During the sub-threshold illumination, the transmembrane potentials of atrial myocytes in all three tissues were gradually forced to approximately -50 mV (the plateau potential observed under illumination, as shown in Figure 2), and these potentials slowly returned to the resting levels upon the termination of illumination.

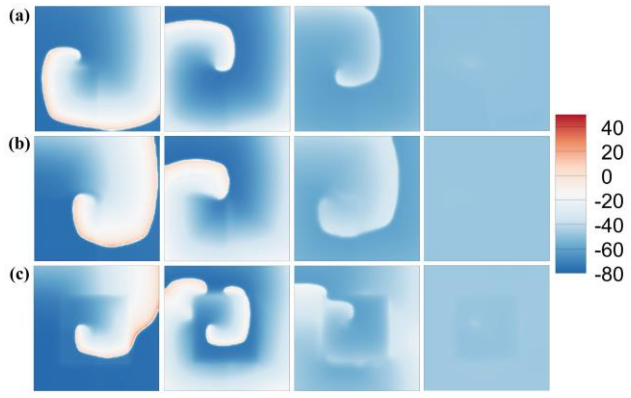

Figure 3. Representative snapshots of spiral waves in 2D sub-threshold illuminating simulations of (a) pure atrial myocytes, (b) atrial diffuse fibrosis and (c) atrial focal fibrosis.

4. Discussion

This study preliminarily explored the impact of sub-threshold illumination on spiral wave dynamics in pure atrial muscle tissue, atrial diffuse fibrosis and focal fibrosis tissues transduced with GtACR1 under chronic AF conditions. At the cellular level, atrial myocytes coupled with Fbs during AF exhibited the shortest APD_{80} after electrical stimulation, with both resting membrane potential and peak membrane potential significantly decreased. During sub-threshold illumination, the transmembrane potential of atrial myocytes was forced at the GtACR1 reversal potential, and cardiomyocytes coupled with Fbs exhibited the slowest potential rise and recovery rates. At the 2D level, spiral waves persisted in pure atrial muscle tissue and atrial diffuse fibrosis tissue, but fragmented in focal fibrosis tissue. However, sub-threshold illumination terminated spiral waves in all three types of tissues.

Previous studies investigating the electrophysiological remodeling at the cellular level within fibrotic regions under chronic AF conditions had not incorporated a Fb electrophysiological model [4, 9]. Instead, they relied solely on the AP model of atrial myocytes in chronic AF [6, 7], further describing the electrophysiological alterations in the fibrotic region's AP model as a 50% reduction in both inward rectifier potassium current (I_{K1}) and L-type calcium channel current (I_{CaL}), alongside a 40% decrease in sodium current (I_{Na}) [10, 11]. These modifications led to outcomes such as prolonged APD, as well as decreased resting and peak transmembrane potentials. The present study, however, integrated the Fb electrophysiological model, revealing that APD_{80} was shortened, with APD_{90} largely aligning with that of non-fibrotic atrial myocytes. The changes in resting and peak potentials concurred with previous findings. Additionally, regarding the alterations in the AP of atrial myocytes transduced with GtACR1 during illumination, our results echoed prior research, demonstrating the "light-induced voltage forcing" mechanism mediated by GtACR1-mediated photocurrents on cardiomyocytes [12-14].

Ochs et al. successfully demonstrated that sub-threshold illumination based on GtACR1 could achieve atrial optogenetic defibrillation, with the required light energy as low as 0.005 mW/mm² [4]. In our study, we employed even lower energy illumination (0.001mW/mm2) and found it to be effective in terminating spiral waves in three types of atrial tissues. However, simulations conducted by Ochs et al. based on three individualized atrial models indicated that this level of light energy was insufficient to successfully terminate AF. We attributed this discrepancy to factors such as the proportion of fibrotic regions, the pattern of fibrosis distribution, and whether or not a Fb electrophysiological

model was incorporated. We aimed to further investigate the energy range of sub-threshold illumination for defibrillation via GtACR1 in future 3D individualized atrial simulation studies.

Hussaini et al. utilized a 2D computational model of ChR2-transduced ventricular tissue from adult mice to simulate the dynamic control of spiral waves by sub-threshold illumination of varying gradients, frequencies, and pulse widths [15-17]. In a parallel study, Majumder et al. employed a 2D model of ChR2-transduced monolayer tissue within the atrial wall from neonatal mice and observed that higher-intensity sub-threshold illumination led to the disruption of spiral waves [18]. Building upon these findings, our study examined a 2D model of GtACR1-transduced human atrial myocardium, incorporating two types of fibrosis. We corroborated the advantages of GtACR1 in terms of the required subthreshold illumination intensity and its efficiency in terminating spiral waves. The model will be further developed to rigorously investigate the impact of various lighting strategies on spiral wave dynamics.

5. Summary

We demonstrated the effects of sub-threshold illumination on cellular electrophysiology and spiral wave dynamics using GtACR1-transduced human atrial cells and 2D model in chronic AF. Under sub-threshold illumination conditions, I_{GtACR1} functioned like the "optogenetic voltage forcing" mechanism observed during normal threshold illumination, preventing re-excitation of atrial myocytes. Consequently, spiral waves were terminated in both pure atrial muscle tissue, as well as in tissues exhibiting diffuse and focal atrial fibrosis.

Acknowledgments

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