

# A Computational Framework for Simulating Cardiac Optogenetics

Patrick M Boyle<sup>1</sup>, John C Williams<sup>2</sup>, Emilia Entcheva<sup>2</sup>, Natalia A Trayanova<sup>1</sup>

<sup>1</sup>Johns Hopkins University, Baltimore, Maryland, USA

<sup>2</sup>Stony Brook University, Stony Brook, New York, USA

## Abstract

*Recent experimental studies have shown that cardiac tissue can be engineered to respond to optical stimulation using Channelrhodopsin-2 (ChR2), a light-gated cation channel. We present the first comprehensive multiscale framework for simulating cardiac optogenetics, following illumination effects from membrane proteins to ventricular contraction. Virtual optogenetic therapy is applied in ventricular models to investigate the design of efficient optical stimulation schemes. Major determinants of threshold irradiance levels are characterized and the Purkinje system is identified as a novel target for optical pacemaking. This study describes a tremendous step forward in our ability to leverage computational tools in the design and optimization of conceptually new optogenetic actuation techniques.*

## 1. Introduction

Cardiac optogenetics is a promising new avenue for altering the behavior of the heart using light instead of electrical current. Channelrhodopsin-2 (ChR2) is a photosensitive cation channel that has been successfully used to inscribe optical sensitivity in cardiac cells, either by direct gene delivery (GD) via lentiviral vector [1] or by coculturing cardiomyocytes with ChR2-rich inexcitable cells (CD) [2]. Experiments have demonstrated the feasibility of using light to control the heart in vitro and in vivo [3]. Computational modeling of this exciting new technology will facilitate the design of efficient and effective optical solutions for controlling cardiac behavior; this will provide a platform for systematically optimizing the delivery of light-sensitive material and the efficiency of stimulation by illumination.

In this study, we present a comprehensive framework for modeling optogenetics in the heart, from single channels to the contracting heart. We exploit the flexibility of the simulation platform to model a variety of optogenetic control schemes for ventricular stimulation, from ventricular pacing to selective optical control of conductive tissue and light-driven contractions that mimic sinus rhythm. Our

analysis reveals key factors for optimizing optical stimulation efficiency and suggests new optogenetic therapy targets.

## 2. Methods

We used a 4-state Markovian model of light-sensitive current ( $I_{ChR2}$ ) [4] that was modified based on patch clamp experiments. Optogenetically-engineered cardiac tissue was modeled as a syncytium of normal myocytes and cells expressing  $I_{ChR2}$ . In gene delivery (GD) mode,  $I_{ChR2}$  was added to existing models of human [5], rabbit [6], or canine [7] ventricular myocytes or rabbit Purkinje fibers [8]. Fig. 1A shows a schematic for GD with generic inward and outward currents. In cell delivery (CD) mode,  $I_{ChR2}$  was added to inexcitable cells with a higher resting potential than myocytes ( $-40\text{ mV}$ ); this donor configuration models the in vitro tandem cell unit approach, where ChR2-rich HEK cells form gap junctions when co-cultured with cardiac cells [2]. Fig. 1C shows characteristic  $I_{ChR2}$  responses, with well-defined peak and plateau phases, in a human model [5] clamped to resting membrane voltage ( $V_m$ ) and illuminated with blue light ( $470\text{ nm}$ ) at several irradiance values.

In experimentally engineered cardiac tissue, donor cells tend to aggregate in a complex patchwork [2, 9]. We distributed ChR2-containing cells in target tissue regions using a two-parameter stochastic algorithm [10]. For each unique delivery configuration,  $D$  and  $P$  controlled the density and patchiness of light-sensitive cells, respectively; as illustrated in Fig. 2A, higher  $P$  resulted in more diffuse distributions. In all cases, clusters of cells containing optogenetic material were seeded from the endocardial surface.

Light does not penetrate tissue without loss of intensity; as a representation of photon scattering effects in optogenetic stimulation, we assumed uniform irradiance at the illuminated surface ( $\bar{E}_e$ ) and modeled attenuation in depth using a simple monoexponential decay. At each point,  $E_e(r) = \bar{E}_e e^{-r/\delta}$ , where  $r$  is the distance to the nearest illuminated node. The wavelength-dependent term  $\delta$  was set to  $570\text{ }\mu\text{m}$ , which produced a good representation

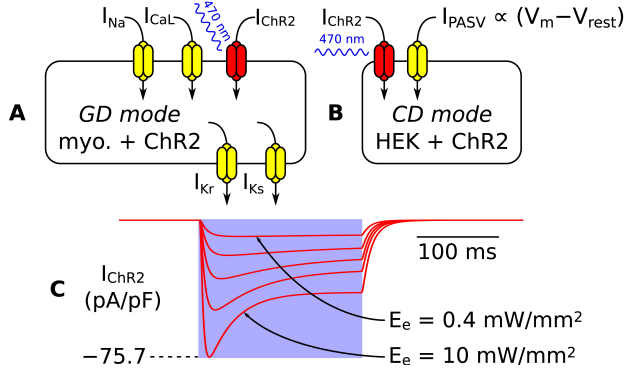


Figure 1. Light-sensitive cells are modeled by adding  $I_{ChR2}$  to either normal cardiomyocytes in gene delivery (GD) mode (A) or nonexcitable donor cells in cell delivery (CD) mode (B); nonexcitable cells are drawn towards a fixed resting potential ( $V_{rest}$ ) by a passive current ( $I_{PASV}$ ). C: With  $V_m$  clamped to  $-85.7 mV$ ,  $I_{ChR2}$  is shown for 200 ms of illumination with several irradiance ( $E_e$ ) values.

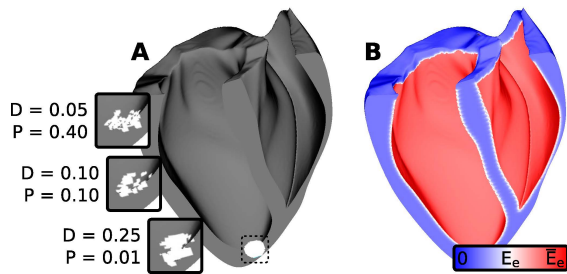


Figure 2. A: Optogenetic material (white) is delivered to a 1 cm-diameter region in the LV apex (dashed line); inset panels show stochastic distributions of ChR2-expressing cells for different combinations of density ( $D$ ) and patchiness ( $P$ ). B: When the endocardium is illuminated (red), effective irradiance ( $E_e$ ) decreases exponentially with depth.

of illumination at 488 nm in optical mapping simulations [11].

Whole-heart optogenetic control schemes were modeled in several biophysically-detailed models validated in previous studies [12–14]. In the MRI-based human model, ChR2-rich cells were delivered to a 1 cm-diameter region in the LV apex; basic features of optical stimulation and significant effects of ChR2 delivery method, spatial distribution, and photon scattering were explored. In the rabbit ventricles with Purkinje system (PS), we investigated potential benefits of cell-specific optogenetic therapy. Optical pacing was applied either to key ventricular sites near PS endpoints or to the entire PS. Finally, in an electromechanical canine model, we assessed hemodynamic function for an optogenetic scheme that mimicked

sinus rhythm.

Electrical activity was simulated in the CARP package [15] using the monodomain formulation for propagation in cardiac tissue. Mechanical activity was simulated using software tools described previously [16].

### 3. Results

The response of the human ventricles to optical stimulation at the LV apex is shown in Fig. 3A. Optogenetic engineering was applied to a 1 cm-diameter hemisphere; in this example,  $I_{ChR2}$  was delivered in CD mode with  $D = 0.25$  and  $P = 0.01$ , producing a large, consolidated light-sensitive region. Illumination was applied from  $t = 0$  to 10 ms, delivering  $2.46 mW/mm^2$  uniformly to the endocardium; this stimulus was just above threshold. Excitation initiated at the right side of the site of tissue engineering ( $t = 40 ms$ ) then activated the rest of the myocardium; Electrotonic effects during activation depolarized CD tissue to  $V_m \approx -15 mV$ .

Figs. 3B-E emphasize key findings from the human model. First, incorporating light attenuation increased threshold irradiance (compare  $E_{e,thr}$  values inside and outside parentheses) by 60% or more. Second,  $E_{e,thr}$  for diffuse ChR2 distribution in GD mode was 33% higher compared to the consolidated pattern; in contrast, for the same preparations in CD mode,  $E_{e,thr}$  was 52% lower. Third, high-density ChR2 regions corresponded to the earliest and latest local activation sites for GD and CD modes, respectively; early excitations in CD mode originated from areas of intermingled ChR2 cells and myocytes.

Fig. 4 compares two possible optogenetic stimulation configurations in the rabbit ventricles with PS. In panel A, GD mode ChR2 was delivered at 10 ventricular locations corresponding to sinus activation sites (6 LV sites: anterior base and apex, posterior base and apex, upper and lower septum; 4 RV sites: anterior, posterior, free wall, and apex); in panel B, the PS was constructed entirely from light-sensitive Purkinje fibers, which were subjected to unattenuated illumination. While the overall activation patterns for the two cases were very similar, the stimulation threshold for the optogenetic PS case was  $> 85\%$  lower.

Finally, simulations in the canine electromechanical model compared the hemodynamic response during sinus rhythm to an optogenetic configuration similar to Fig. 4A. As shown in Fig. 5A, electrical activation for the last in a series of 40 simulated beats emanated from illuminated tissue regions following a 10 ms light pulse, leading to a vigorous ventricular contraction. Inspection of spatial and temporal profiles in strain with respect to end diastolic state (Figs. 5 B & C) revealed early myofilament shortening in the septum and the RV free wall, near optical stimulation sites, followed by contraction in the later-activating LV free wall. All optogenetic sites were stimu-

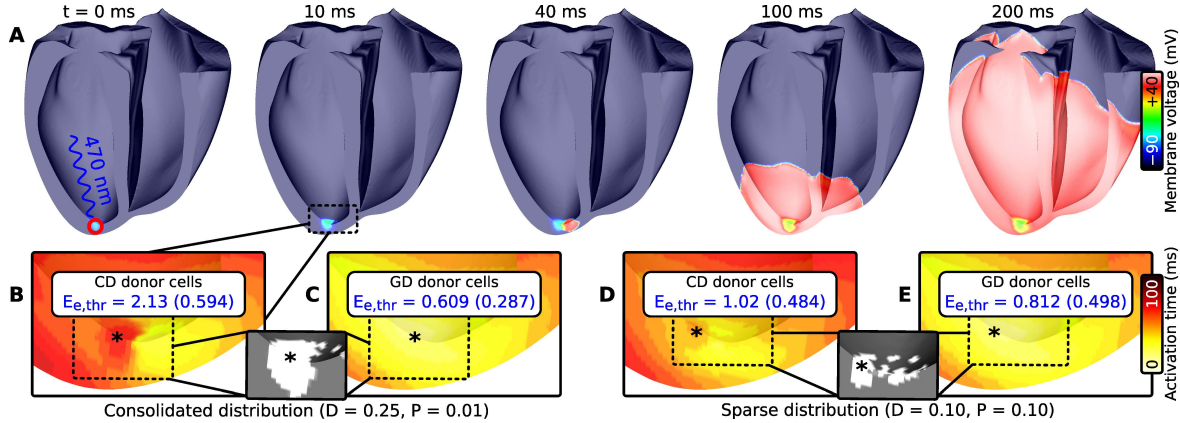


Figure 3. **A:** Simulated human ventricular response to 10 ms blue light pulse at LV apex (CD mode,  $D = 0.25$ ,  $P = 0.01$ ). **B & C:** Activation sequences for CD and GD modes with threshold irradiances in  $mW/mm^2$ ; values in parentheses indicate reduced thresholds due to omission of photon scattering effects. Asterisks (\*) highlight locations with high ChR2 density. **D & E:** Same as above but with a more diffuse spatial distribution of ChR2-rich cells.

lated simultaneously at the moment of illumination, eliminating the detailed natural spatiotemporal activation sequence; nonetheless, LV and RV pressure-volume loops for optical pacing (Fig. 5D) closely matched those generated during sinus rhythm.

#### 4. Discussion and conclusions

In this study, we present the first realistic simulations of optogenetics in biophysically-detailed models of the heart. Engineering cardiac tissue to respond reliably and efficiently to optical stimulation is associated with many practical challenges; we incorporate key aspects of the cell delivery and illumination processes as a proof-of-concept method for investigating these challenges. Our findings demonstrate that several such factors, especially light attenuation and spatial distribution of ChR2-expressing cells, play an important role in determining threshold irradiance levels for tissue excitation.

In vitro studies of cardiac optogenetics have yielded promising results for both CD [2] and GD [1] modes, but neither has emerged as the preferred alternative. Fig. 3 emphasizes that the distribution of ChR2 in engineered tissue will be a key factor in the settling of this question. It is clear that more diffuse distributions of engineered cells reduce irradiance threshold for CD preparations and have the opposite effect for GD mode. If large, consolidated ChR2 expression patterns prove difficult to produce in vivo, CD may prove to be the superior option.

Our findings identify the Purkinje fibers as an intriguing target for optogenetic stimulation. Preliminary efforts to deliver genes by lentivirus to specific cell populations have been successful [17], which suggests it may be feasible to selectively enhance the PS with ChR2. Fig. 4 suggests that

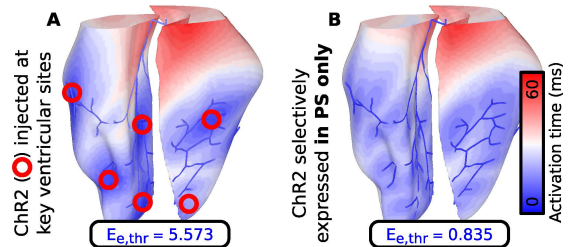


Figure 4. Comparison of endocardial response to 2 ms light pulse in rabbit heart with GD-mode ChR2 expressed in either (A) 2 mm-diameter key ventricular sites near Purkinje system (PS) endpoints or (B) the entire PS.

energy requirements for the resulting optogenetic control scheme would be dramatically reduced compared to stimulating regions of engineered ventricular tissue. This could be a consequence of restrictive PS geometry effects on local source-sink relationships.

Insights on the determinants of optical excitability and exciting new pacemaking concepts are the first of many possibilities for using simulations to explore cardiac optogenetics. As Fig. 5 shows, we have developed the first comprehensive computational toolkit for investigating these phenomena at every level of the cardiac response, including hemodynamic function. With continuing work in this area, we are confident that biophysical modeling will play an important role in the design of efficient and effective strategies for optogenetic control in the heart.

#### Acknowledgements

Dr. Boyle is supported by a fellowship from the Natural Sciences and Engineering Research Council of Canada. This research was also supported by National Institutes of

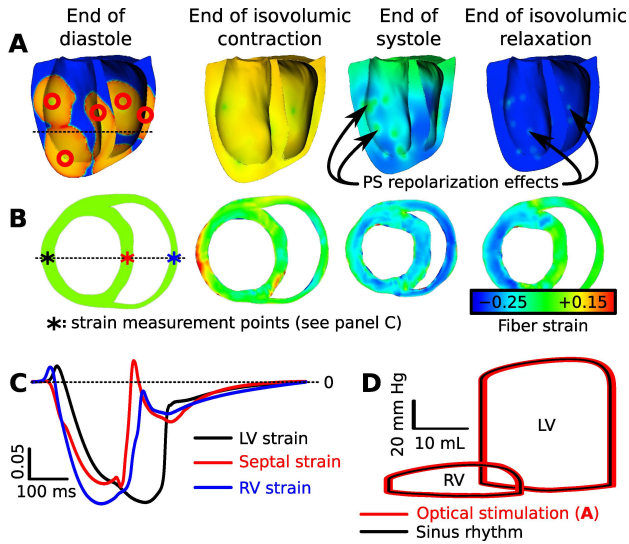


Figure 5. **A & B:** Long-axis  $V_m$  (same scale as Fig. 3A) and short-axis strain profiles at key instants during the cardiac cycle illustrate the electromechanical response of the canine ventricles to optical stimulation of key ventricular sites; PS fibers were simulated but could not be rendered; strain is calculated with respect to the end diastolic state. **C:** Strain at select points for the entire heartbeat. **D:** Pressure-volume loops for the optical response show a similar hemodynamic response to sinus rhythm.

Health grants R01HL111649 and R01HL103428 and by National Science Foundation grant CBET0933029.

## References

- [1] Abilez OJ, Wong J, Prakash R, Deisseroth K, Zarins CK, Kuhl E. Multiscale computational models for optogenetic control of cardiac function. *Biophys J* Sep 2011; 101(6):1326–1334.
- [2] Jia Z, Valiunas V, Lu Z, Bien H, Liu H, Wang HZ, Rosati B, Brink PR, Cohen IS, Entcheva E. Stimulating cardiac muscle by light: cardiac optogenetics by cell delivery. *Circ Arrhythm Electrophysiol* Oct 2011;4(5):753–760.
- [3] Bruegmann T, Malan D, Hesse M, Beiert T, Fuegeman CJ, Fleischmann BK, Sasse P. Optogenetic control of heart muscle in vitro and in vivo. *Nat Methods* Nov 2010; 7(11):897–900.
- [4] Nikolic K, Grossman N, Grubb MS, Burrone J, Toumazou C, Degenaar P. Photocycles of channelrhodopsin-2. *Photochem Photobiol* 2009;85(1):400–411.
- [5] ten Tusscher KHWJ, Panfilov AV. Alternans and spiral breakup in a human ventricular tissue model. *Am J Physiol Heart Circ Physiol* Sep 2006;291(3):H1088–H1100.
- [6] Mahajan A, Shiferaw Y, Sato D, Baher A, Olcese R, Xie LH, Yang MJ, Chen PS, Restrepo JG, Karma A, Garfinkel A, Qu Z, Weiss JN. A rabbit ventricular action potential model replicating cardiac dynamics at rapid heart rates. *Biophys J* Jan 2008;94(2):392–410.
- [7] Hund TJ, Rudy Y. Rate dependence and regulation of action potential and calcium transient in a canine cardiac ventricular cell model. *Circulation* Nov 2004;110(20):3168–3174.
- [8] Aslanidi OV, Sleiman RN, Boyett MR, Hancox JC, Zhang H. Ionic mechanisms for electrical heterogeneity between rabbit purkinje fiber and ventricular cells. *Biophys J* Jun 2010;98(11):2420–2431.
- [9] Rosen AB, Kelly DJ, Schuldt AJT, Lu J, Potapova IA, Doronin SV, Robichaud KJ, Robinson RB, Rosen MR, Brink PR, Gaudette GR, Cohen IS. Finding fluorescent needles in the cardiac haystack: tracking human mesenchymal stem cells labeled with quantum dots for quantitative in vivo three-dimensional fluorescence analysis. *Stem Cells* Aug 2007;25(8):2128–2138.
- [10] Comtois P, Nattel S. Interactions between cardiac fibrosis spatial pattern and ionic remodeling on electrical wave propagation. *Conf Proc IEEE Eng Med Biol Soc* 2011; 2011:4669–4672.
- [11] Bishop MJ, Rodriguez B, Eason J, Whiteley JP, Trayanova N, Gavaghan DJ. Synthesis of voltage-sensitive optical signals: application to panoramic optical mapping. *Biophys J* Apr 2006;90(8):2938–2945.
- [12] Moreno JD, Zhu ZI, Yang PC, Bankston JR, Jeng MT, Kang C, Wang L, Bayer JD, Christini DJ, Trayanova NA, Ripplinger CM, Kass RS, Clancy CE. A computational model to predict the effects of class I anti-arrhythmic drugs on ventricular rhythms. *Sci Transl Med* Aug 2011;3(98):98ra83.
- [13] Boyle PM, Deo M, Plank G, Vigmond EJ. Purkinje-mediated effects in the response of quiescent ventricles to defibrillation shocks. *Ann Biomed Eng* Feb 2010; 38(2):456–468.
- [14] Provost J, Gurev V, Trayanova N, Konofagou EE. Mapping of cardiac electrical activation with electromechanical wave imaging: an in silico-in vivo reciprocity study. *Heart Rhythm* May 2011;8(5):752–759.
- [15] Vigmond EJ, dos Santos RW, Prassl AJ, Deo M, Plank G. Solvers for the cardiac bidomain equations. *Prog Biophys Mol Biol* 2008;96(1-3):3–18.
- [16] Gurev V, Lee T, Constantino J, Arevalo H, Trayanova NA. Models of cardiac electromechanics based on individual hearts imaging data: image-based electromechanical models of the heart. *Biomech Model Mechanobiol* Jun 2011; 10(3):295–306.
- [17] Zhang XY, Kutner RH, Bialkowska A, Marino MP, Klimstra WB, Reiser J. Cell-specific targeting of lentiviral vectors mediated by fusion proteins derived from sindbis virus, vesicular stomatitis virus, or avian sarcoma/leukosis virus. *Retrovirology* 2010;7:3.

Address for correspondence:

Patrick M Boyle  
 Institute for Computational Medicine  
 316 Hackerman Hall  
 3400 North Charles Street  
 Baltimore MD 21218 USA  
 pmjboyle@jhu.edu