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Computational optogenetics: A novel continuum framework for the photoelectrochemistry of living systems

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Abstract
Electrical stimulation is currently the gold standard treatment for heart rhythm disorders. However, electrical pacing is associated with technical limitations and unavoidable potential complications. Recent developments now enable the stimulation of mammalian cells with light using a novel technology known as optogenetics. The optical stimulation of genetically engineered cells has significantly changed our understanding of electrically excitable tissues, paving the way towards controlling heart rhythm disorders by means of photostimulation. Controlling these disorders, in turn, restores coordinated force generation to avoid sudden cardiac death. Here, we report a novel continuum framework for the photoelectrochemistry of living systems that allows us to decipher the mechanisms by which this technology regulates the electrical and mechanical function of the heart. Using a modular multiscale approach, we introduce a non-selective cation channel, channelrhodopsin-2, into a conventional cardiac muscle cell model via an additional photocurrent governed by a light-sensitive gating variable. Upon optical stimulation, this channel opens and allows sodium ions to enter the cell, inducing electrical activation. In side-by-side comparisons with conventional heart muscle cells, we show that photostimulation directly increases the sodium concentration, which indirectly decreases the potassium concentration in the cell, while all other characteristics of the cell remain virtually unchanged. We integrate our model cells into a continuum model for excitable tissue using a nonlinear parabolic second order partial differential equation, which we discretize in time using finite differences and in space using finite elements. To illustrate the potential of this computational tool box, we virtually inject our photosensitive cells into different locations of a human heart, and explore its activation sequences upon photostimulation. Our computational optogenetics tool box allows us to virtually probe landscapes of process parameters, and to identify optimal photostimulation sequences with the goal to pace human hearts with light and, ultimately, to restore mechanical function.

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1. Motivation

The human heart propels over 7000 l of blood through the body daily, beating more than 40 million times a year. It is a remarkably efficient, durable, and reliable mechanical pump, precisely regulated by spatially and temporally varying electrical and chemical fields. Disturbed conduction and uncoordinated electrical signals can induce abnormal heart
rhythms, which may critically reduce mechanical function (Dubin, 2000; Tsamis et al., 2011). A delicate balance between
the electrical and chemical gradients across the cell membrane governs the electrophysiological activity of excitable
cardiac cells. To maintain these gradients, the cell membrane is selectively permeable with respect to different ions at
different points in time (Berne and Levy, 2001). In a cardiac muscle cell, for example, the potential difference across the cell
membrane is approximately −80 mV at rest, meaning the cell is negatively charged. It can be excited by an external
electrical stimulus that pushes its transmembrane potential beyond a critical threshold value. This triggers the influx of
sodium ions, generating a rapid depolarization, and the transmembrane potential increases to +20 mV. Over time, the
coordinated interplay between sodium, potassium, and calcium ions brings the transmembrane potential smoothly back to
its resting state (Keener and Sneyd, 2004).

Since the late 1950s, permanent cardiac pacemakers have been used successfully to correct rhythm disorders and
restore smoothly propagating waves of electrical excitation (Furman and Schwedel, 1959). Pacemakers deliver electrical
stimuli via implanted pacing leads that are in contact with the heart muscle. Although permanent pacemaker implantation
is a standard procedure today, pacing lead placement continues to be a subject of great controversy (Modi et al., 2011).
Choosing the optimal pacing site is becoming more challenging (Puwal and Roth, 2009), in particular in view of novel
stimulation strategies such as single chamber, dual chamber, and biventricular pacing. A potential complication of lead
placement is cardiac perforation, which can have severe clinical consequences including pericardial effusion, cardiac
tamponade, pneumothorax, and death (Mahapatra et al., 2005). However, to date, there is no conceptual alternative to
electrical pacing that avoids direct contact stimulus delivery (Khan et al., 2005).

For more than 40 years, biologists have studied microorganisms that produce proteins to directly regulate the flow of
charged ions across their plasma membrane in response to light (Oesterhelt and Stoeckenius, 1971). The first identified
protein of this kind was the light-gated ion pump bacteriorhodopsin, transporting positively charged hydrogen ions across
the cell membrane (Oesterhelt and Stoeckenius, 1971). While bacteriorhodopsin naturally acts as an on-switch for
electrically active cells, the second reported light-gated ion pump, halorhodopsin, transports negatively charged chloride
ions, thereby acting as an off-switch (Matsuno-Yagi and Mukohata, 1977). Despite intense research (Veselodov, 1998),
the first reported light-gated ionic channel, channelrhodopsin, was only identified 10 years ago (Nagel et al., 2002), but has
since then revolutionized neuroscience. Channelrhodopsin consists of seven transmembrane proteins embedded in the
lipid bilayer (Feng and Klug, 2006) and absorbs blue light through its interaction with retinal, see Fig. 1.

Since the early 1990s, we have known that phototaxis and photophobic responses in the green alga Chlamydomonas reinhardtii are mediated by rhodopsins with a microbial-type all-trans retinal chromophore (Hegemann et al., 1991). The photochemical isomerization of this all-trans retinal to 13-cis retinal is illustrated in Fig. 2. It occurs at peak absorption wavelengths of 470 nm, opening channelrhodopsin non-specifically to sodium, potassium, and calcium cations in response to blue light. In the dark, the covalently bound retinal spontaneously relaxes to all-trans, providing closure of the channel and regeneration of the chromophore. In 2005, channelrhodopsin was first introduced genetically using engineered viruses (Boyden et al., 2005), a technique that is now widely known as optogenetics (Deisseroth, 2011). Since then, optical tools to control the electrical activity of nerve cells have rapidly evolved (Szobota and Isacoff, 2010), and are now gaining widespread use in neuronal research and medicine (Hegemann and Möglich, 2011). While initial applications of optogenetics have been restricted exclusively to the neuronal system, optogenetic tools have now advanced to a level of maturity where they can confidently be applied to other cells and organs. Natural first candidates are stem cells, electrically active glial cells, muscle cells, and cardiac cells (Arrenberg et al., 2010; Jia et al., 2011). Since light can be delivered at a distance, optical pacing has the potential to become a more precise and less invasive alternative to existing electrical pacing strategies (Abilez et al., 2011).

![Fig. 1. Channelrhodopsin-2 is a light-gated cation channel native to the green alga Chlamydomonas reinhardtii. It consists of seven transmembrane proteins and absorbs blue light through its interaction with retinal. Photoisomerization of retinal opens the channel to sodium ions, which have a higher concentration outside than inside the cell. To make our cells responsive to light, and allow sodium concentrations to equilibrate, we induce channelrhodopsin into a conventional cardiac muscle cell model.](image_url)

To model the light sensitivity of channelrhodopsin, we adopt the kinetics of a three-state photocycle model (Hegemann et al., 2005; Nikolic et al., 2009), which consists of a closed, photosensitive, excitable state $g_{\text{closed}}$, an open state $g_{\text{ChR2}}$, and a closed, desensitized, absolutely refractory state $g_{\text{refrac}}$ (Nagel et al., 2003), as illustrated in Fig. 4. Upon photo absorption, molecules which are in the closed state $g_{\text{closed}}$ undergo a fast transition into the open state $g_{\text{ChR2}}$. After being open for some time, molecules spontaneously transition into the absolutely refractory state $g_{\text{refrac}}$, where the ion channels are closed, but the molecules are not yet ready to photoswitch again. After the refractory period, the molecules finally return to the closed state $g_{\text{closed}}$, ready to undergo a new photocycle when exposed to light (Nikolic et al., 2006). Fig. 4 suggests the following first order model for the channelrhodopsin photocycle:

$$g_{\text{ChR2}} = \epsilon m_{\text{pho}} g_{\text{closed}} - \Gamma, g_{\text{ChR2}}$$

$$g_{\text{refrac}} = \Gamma, g_{\text{ChR2}} - \Gamma, g_{\text{refrac}}$$

The objective of this manuscript is to establish a novel continuum model for the photoelectrochemistry of living systems that will allow us to virtually explore the potential of optogenetic pacing. The rationale for creating such a model is that it will enable patient-specific predictions of ion channel dynamics, ionic concentrations, and action potential profiles across the heart, which are outside the reach of experimental measurements in humans. Fig. 3 illustrates the underlying approach in which the different physical fields interact across the different scales: on the molecular level, optical stimulation opens the cation channel channelrhodopsin initiating a photocurrent. On the subcellular level, this photocurrent increases the chemical concentration of sodium ions inside the cell. On the cellular level, concentration changes evoke changes in the electrical potential and excite the cell. On the tissue level, changes in the electrical potential propagate across the system in the form of smooth excitation waves.

In Section 2, we summarize the mathematical model for the channelrhodopsin photocycle and illustrate how channelrhodopsin can be introduced into a conventional cardiac cell model via an additional photocurrent governed by a light-sensitive gating variable. We characterize this additional photocurrent in Section 3, and demonstrate its impact on the intracellular ion concentrations in Section 4, and on the transmembrane potential in Section 5. In a side-by-side comparison with conventional cardiac muscle cells, we perform virtual case studies to demonstrate the features of genetically engineered light-sensitive cells in terms of gating variables, ionic currents, ion concentrations, and transmembrane potentials. Since our baseline model for conventional cardiac cells is well-characterized and fairly standard, we summarize its equations in the Appendix. We proceed by embedding these single cells into a novel continuum framework for photoelectrochemistry to simulate the spatial and temporal variation of electrical and chemical fields across the human heart. We briefly summarize the finite element discretization of the governing equations in Section 6. In Section 7, we virtually inject our light-sensitive cells into different locations of a human heart and explore its response to photostimulation. In Section 8, we conclude by discussing computational optogenetics and the potential of turning light into force in the context of pacing hearts with light.
Variables as illustrated in Fig. 5 where $G$ is the quantum efficiency of the channelrhodopsin system, and $n_{\text{pho}}$ is the number of photons hitting the cell per second. Here, we choose $\Gamma_\epsilon = 0.06129$, $\Gamma_c = 0.00422$, and $n_{\text{pho}} = 0.018$. Herein, $g_{\text{ChR2}}$, $g_{\text{refrac}}$, and $g_{\text{closed}}$ define the fraction of molecules in the open, refractory, and closed states. Initially, all molecules are in the closed state, i.e., $g_{\text{closed}} = 1$, $g_{\text{ChR2}} = 0$, and $g_{\text{refrac}} = 0$. Since all three fractions sum up to one, i.e., $g_{\text{ChR2}} + g_{\text{refrac}} + g_{\text{closed}} = 1$, the photocycle system (1) can be characterized through two independent variables (Ishizuka et al., 2006), e.g., the fraction of molecules in the open and in the refractory states

$$g_{\text{closed}} = \Gamma_\epsilon \frac{\Gamma_c}{\text{refrac}} - en_{\text{pho}} g_{\text{closed}}$$

(1)

$\Gamma_\epsilon$ and $\Gamma_c$ are the rates of recovery and full closure, $\epsilon$ is the quantum efficiency of the channelrhodopsin system, and $n_{\text{pho}}$ is the number of photons hitting the cell per second. Here, we choose $\Gamma_\epsilon = 0.06129$, $\Gamma_c = 0.00422$, and $n_{\text{pho}} = 0.018$. Herein, $g_{\text{ChR2}}$, $g_{\text{refrac}}$, and $g_{\text{closed}}$ define the fraction of molecules in the open, refractory, and closed states. Initially, all molecules are in the closed state, i.e., $g_{\text{closed}} = 1$, $g_{\text{ChR2}} = 0$, and $g_{\text{refrac}} = 0$. Since all three fractions sum up to one, i.e., $g_{\text{ChR2}} + g_{\text{refrac}} + g_{\text{closed}} = 1$, the photocycle system (1) can be characterized through two independent variables (Ishizuka et al., 2006), e.g., the fraction of molecules in the open and in the refractory states

$$g_{\text{ChR2}} = n_{\text{pho}} - [en_{\text{pho}} + \Gamma_\epsilon] g_{\text{ChR2}} - en_{\text{pho}} g_{\text{refrac}}$$

(2)

$$g_{\text{refrac}} = \Gamma_\epsilon g_{\text{ChR2}} - \Gamma_c g_{\text{refrac}}$$

(3)

We identify the state $g_{\text{ChR2}}$ as the channelrhodopsin gating variable and integrate it into a well-characterized ventricular cell model (ten Tusscher et al., 2004; Wong et al., 2011) characterized through a total of $n_{\text{gate}} = 14$ gating variables as illustrated in Fig. 5

$$g_{\text{gate}} = [g_m, g_h, g_f, g_{\text{ChR2}}, g_{\text{x1}}, g_{\text{x2}}, g_{\text{Kr}}, g_{\text{Cat}}, g_{\text{Na}}, g_{\text{K1}}, g_{\text{Ca}}, g_{\text{Ks}}, g_{\text{pCa}}, g_{\text{pK}}, g_{\text{refrac}}, g_{\text{closed}}, g_{\text{ChR2}}]$$

In particular, these are the sodium activation gate $g_m$, the fast and slow sodium inactivation gates $g_h$ and $g_j$, the channelrhodopsin activation gate $g_{\text{ChR2}}$, the rapid delayed rectifier activation and inactivation gates $g_{xr1}$ and $g_{xr2}$, the slow delayed rectifier activation gate $g_{xs}$, the transient potassium activation and inactivation gates $g_r$ and $g_s$, the long-lasting L-type calcium channel activation and inactivation gates $g_d$, $g_f$, and $g_{fCa}$, and the calcium release activation gate $g_g$. The gating variables are parameterized in terms of the transmembrane potential $\phi$, the ionic concentrations $c_\text{ion}$, and the gating variables $g_{\text{gate}}$ themselves. Their evolution is governed by classic Hodgkin–Huxley type equations

$$g_{\text{gate}} = \frac{1}{t_{\text{gate}}(\phi)} [g_{\text{gate}}(\phi,c_\text{ion}) - g_{\text{gate}}]$$

(4)

each characterized through a steady state value $g_{\text{gate}}^\infty$ and a time constant $t_{\text{gate}}$ for reaching this steady state (Wong et al., 2011). Both are usually exponential functions of the transmembrane potential $\phi$, see Appendix for details. Fig. 6 illustrates the temporal evolution of all $n_{\text{gate}} = 14$ gating variables for the conventional cell, electrically stimulated for 1 ms, dashed

Here, we choose \(c_{Na}^{f} = 153 \text{ mM} \) (Ishizuka et al., 2006) and \(c_{Na}^{i} = 11.6 \text{ mM} \), calibrated to match the experimental reversal potential of channelrhodopsin, and \(e_{Na}^{f} = 140 \text{ mM} \), \(e_{Na}^{i} = 11.6 \text{ mM} \), and \(\phi_{Na,0} = 1.79 \text{ mV} \). Accordingly, \(\phi_{ChR2} = [RT]/[zF] \cdot \log(cNa^{f}/cNa^{i}) + \phi_{Na,0} \). In contrast to our previous approach, where we have photostimulated an atrial nodal cell (Abize et al., 2011), we now integrate the channelrhodopsin photocurrent \(I_{ChR2} \) into our conventional ventricular cell model (ten Tusscher et al., 2004; Wong et al., 2011), defined through a total of \(n_{ch} = 16 \) ionic currents, see Fig. 5.

In particular, these are the fast sodium current \(I_{Na} \), the background sodium current \(I_{bNa} \), the sodium potassium pump current \(I_{NaP} \), the sodium calcium exchanger current \(I_{NaCa} \), the channelrhodopsin photocurrent \(I_{ChR2} \), the inward rectifier current \(I_{IK1} \), the rapid delayed rectifier current \(I_{Kd} \), the slowly delayed rectifier current \(I_{Ks} \), the plateau potassium current \(I_{P} \), the transient outward current \(I_{O} \), the L-type calcium current \(I_{CaL} \), the background calcium current \(I_{CaB} \), the plateau calcium current \(I_{CaP} \), the leakage current \(I_{bCa} \), and the sarcolemal reticulum uptake and release currents \(I_{u} \) and \(I_{rel} \), see Appendix for details.

4. Mathematical model of ionic concentrations

From a chemical point of view, light induces a channelrhodopsin photocurrent \(I_{ChR2} \), which directly impacts the intracellular sodium concentration \(c_{Na} \):

\[
\dot{c}_{Na} = -\frac{1}{FV} [I_{Na} + I_{bNa} + 3I_{NaCa} + 3I_{NaK} + I_{ChR2}]
\]
where $V$ is the cytosolic volume and $F$ is the Faraday constant. The sodium concentration will directly, and indirectly through the resulting changes in the transmembrane potential $\phi$, affect all other ionic concentrations in the cell. The biochemistry of our cell model is characterized through $n_{\text{ion}} = 4$ ion concentrations $c_{\text{Na}}, c_{\text{K}}, c_{\text{Ca}}$, and $c_{\text{Ca}_{\text{SR}}}$, where $c_{\text{Na}}, c_{\text{K}},$ and $c_{\text{Ca}}$ are the intracellular sodium, potassium, and calcium concentrations, and $c_{\text{Ca}_{\text{SR}}}$ is the calcium concentration in the sarcoplasmic reticulum. The concentrations obey evolution equations of the following format:

$$\dot{c}_{\text{ion}} = f_{\text{ion}}(\phi, g_{\text{gate}}, c_{\text{ion}})$$

where $f_{\text{ion}}(\phi, g_{\text{gate}}, c_{\text{ion}})$ is a function that depends on the transmembrane potential $\phi$, the gate current $g_{\text{gate}}$, and the concentration of the ion. The current dynamics for the electrically stimulated cell have been delayed by 34 ms for the purposes of comparison against the optically stimulated cell. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)
variables and ion concentrations locally on the integration point level (Wong et al., 2011). We solve the resulting staggered

accordingly, we introduce the membrane potential as global degree of freedom at each finite element node, and the gating

gap junction channels (Jia et al., 2011). We can characterize its spatio-temporal evolution through a nonlinear parabolic

second-order partial differential equation

\[ c_{\text{ion}} = \frac{q}{C_0} \]

which are parameterized in terms of the transmembrane potential \( \phi \), the gating variables \( g_{\text{gate}} \), and the ion concentrations \( c_{\text{ion}} \) themselves, see Appendix for details. Fig. 8 illustrates the temporal evolution of all \( n_{\text{gate}} = 14 \) gating variables for the conventional cell, electrically stimulated for 1 ms, dashed black lines, and for the genetically engineered cell, light stimulated for 30 ms, solid blue lines. While the calcium concentrations inside the cell \( c_{\text{Ca}} \) and in the sarcoplasmic reticulum \( c_{\text{Ca}}^{\text{SR}} \) remain unaffected by photostimulation, the intracellular sodium concentration \( c_{\text{Na}} \) increases with light. Through the sodium potassium pump current \( I_{\text{NaK}} \), this has a direct impact on the intracellular potassium concentration \( c_{\text{K}} \), which decreases upon photostimulation.

5. Mathematical model of action potential propagation

From an electrical point of view, light induces a channelrhodopsin current \( I_{\text{ChR2}} \), which directly impacts the sodium concentration \( c_{\text{Na}} \), and thus, the action potential propagation \( \phi \) in the heart. Unlike the local ion concentrations for single cells, however, the action potential \( \phi \) is a global field variable (Görttepe and Kuhl, 2009). It propagates from cell to cell via gap junction channels (Jia et al., 2011). We can characterize its spatio-temporal evolution through a nonlinear parabolic second-order partial differential equation

\[ \dot{\phi} = f^\phi(\phi, g_{\text{gate}}, c_{\text{ion}}) + \text{div} \, q(\phi) \]  

driven by a nonlinear local source term \( f^\phi \) at the single cell level, and by a linear global flux term, \( \text{div} \, q \), the divergence of the propagation vector \( q \) at the organ level. We identify the local source term

\[ f^\phi = -\frac{1}{C} \left[ I_{\text{in}} + I_{\text{out}} + I_{\text{Na}} + I_{\text{NaCa}} + I_{\text{ChR2}} + I_{\text{I1}} + I_{\text{K}} + I_{\text{pK}} + I_{\text{f0}} + I_{\text{fCa}} + I_{\text{pCa}} \right] \]

as the negative sum of all transmembrane currents scaled by the cell membrane capacitance per unit surface area \( C \). To account for the nonlocal nature of propagating excitation waves in the heart, we introduce the propagation vector

\[ q = D \cdot \nabla \phi \]

through the second-order diffusion tensor \( D \) scaling the gradient of the action potential field \( \nabla \phi \). Fig. 9 illustrates the five phases of the action potential \( \phi \) for the conventional cell, electrically stimulated for 1 ms, dashed black lines, and for the genetically engineered cell, light stimulated for 30 ms, solid blue lines. We can clearly see the impact of photostimulation, which requires the entire stimulation period of 30 ms to push the cell beyond its excitation threshold. For visualization purposes, we have shifted the transmembrane potential of the electrical stimulation, dashed lines, by 34 ms, which agrees qualitatively with the time delay of 19.7 ± 3.4 ms reported in the literature (Bruegmann et al., 2010). Beyond the stimulation threshold, both cells display an almost identical electrical response with the characteristic rapid upstroke, early, partial repolarization, plateau, final repolarization, and rest phases. Again, this agrees nicely with experimental findings (Bruegmann et al., 2010).

6. Computational model of light-activated cells

To solve the spatio-temporal problem of electrochemical coupling (4), (14) and (15) for the gating variables \( g_{\text{gate}} \), the intracellular ion concentrations \( c_{\text{ion}} \), and the transmembrane potential \( \phi \), we apply a finite difference scheme in time and a finite element scheme in space. Due to the global nature of the membrane potential introduced through the diffusion term \( \text{div} \, q(\phi) \), we suggest a \( C^{1-} \)-continuous finite element interpolation for the membrane potential \( \phi \), while a \( C^{1+} \)-continuous interpolation is sufficient for the sets of gating variables \( g_{\text{gate}} \) and ion concentrations \( c_{\text{ion}} \) (Görttepe and Kuhl, 2009, 2010). Accordingly, we introduce the membrane potential as global degree of freedom at each finite element node, and the gating variables and ion concentrations locally on the integration point level (Wong et al., 2011). We solve the resulting staggered

6.1. Chemical problem—local discretization on the integration point level

We typically initialize the chemical state variables at t₀ with their resting state values. Our goal is to determine the chemical state variables, i.e., the n_gate = 14 gating variables \( \phi \), and the n_ion = 4 ionic currents \( c_{ion} \) at each time step \( t \) for a given action potential \( \phi \) at \( t \), and known chemical state variables from the previous time step \( t_{n-1} \). For the temporal discretization, we partition the time interval of interest \( T \) into \( n_{stp} \) subintervals \([t_{n-1},t_{n}+1] \) as \( T = \bigcup_{n=0}^{n_{stp}-1} [t_{n},t_{n+1}] \) and apply a standard backward Euler time integration scheme in combination with a finite difference approximation of the first-order time derivatives of the gating variables \( \dot{\phi} \) and \( \dot{c}_{ion} \):

\[
\dot{\phi} = \frac{f_{\phi}(\phi, c_{gate}) - f_{\phi}^{\infty}(\phi, c_{gate})}{\Delta t}
\]

\[
\dot{c}_{ion} = \frac{f_{c}(\phi, c_{gate}, c_{ion}) - f_{c}^{\infty}(\phi, c_{gate}, c_{ion})}{\Delta t}
\]

Here and from now on, we omit the index \( (\circ)_{n+1} \) for the sake of clarity, and introduce the common abbreviation \( \Delta t = t - t^n > 0 \) for the linear set of gating equations (4) into a set of update equations for the gating variables \( \phi \) at the current time step \( t \):

\[
\phi_{gate}^{n+1} = \phi_{gate}^{n} + \frac{1}{\tau_{gate}(\phi)} \left[ f_{\phi}(\phi, c_{gate}) - f_{\phi}^{\infty}(\phi, c_{gate}) \right] \Delta t
\]

The gating variables define the n_crit = 16 ionic currents \( I_{crit}(\phi, \phi_{gate}, c_{ion}) \), which alter the n_ion = 4 intracellular ion concentrations \( c_{ion} \). With the help of the finite difference approximation (18), we reformulate the nonlinear set of concentration equations (14) in the following residual format:

\[
R_{ion}^{c} = c_{ion}^{n+1} - c_{ion}^{n} - f_{ion}(\phi, \phi_{gate}, c_{ion}) \Delta t = 0
\]

To obtain the \( \mathbf{K}_{ion}^{C} \) iteration matrix for the local Newton iteration on the integration point level, we linearize the discrete algorithmic residual

\[
\mathbf{K}_{ion}^{C} = d_{c_{ion}} R_{ion}^{c} = 1 - d_{c_{ion}} f_{ion}^{c}
\]

At the end of each Newton iteration, we update the set of ion concentrations \( c_{ion} \), set of gating variables \( \phi_{gate} \), and the set of ionic currents \( I_{crit} \), and continue to iterate locally until we achieve convergence. This local inner loop can be understood as a modern implicit version of the iterative update procedure of the original Rush–Larsen algorithm (Rush and Larsen, 1978).

Fig. 9. Genetically engineered light sensitive cardiac cell stimulated conventionally with an electric field, dashed lines, and optically with light, solid lines. Temporal evolution of transmembrane potential \( \phi \). The characteristic action potential consists of five phases. Phase 0: the rapid upstroke is generated through an influx of positively charged sodium ions. Phase 1: early, partial repolarization is initiated through the efflux of positively charged potassium ions. Phase 2: during the plateau, the net influx of positively charged calcium ions is balanced by the efflux of positively charged potassium ions. Phase 3: final repolarization begins when the efflux of potassium ions exceeds the influx of calcium ions. Phase 4: throughout the interval between end of repolarization and the beginning of the next cycle the cell is at rest. The transmembrane potential for the electrically stimulated cell has been delayed by 34 ms for the purposes of comparison against the optically stimulated cell. This delay agrees nicely with the time delay of activation of 19.7 ± 3.4 ms reported in the literature (Bruegmann et al., 2010). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

6.2. Electrical problem—global discretization on the node point level

To evaluate the action potential \( \phi \) on the node point level, we transform the electrical problem (15) into its residual format

\[
R^0 = \phi - \text{div}(q) - f^0 = 0
\]  

(22)

in the domain of interest \( \Omega \). We complement this initial boundary value problem with the corresponding Dirichlet and Neumann boundary conditions \( \phi = \bar{\phi} \) on \( \partial B_\phi \) and \( \mathbf{q} \cdot \mathbf{n} = \bar{\mathbf{q}} \) on \( \partial B_q \), typically using homogeneous Neumann boundary conditions \( \mathbf{q} \cdot \mathbf{n} = 0 \) on the entire boundary \( \partial B \). As initial conditions, \( \phi_0(x) = \phi(x, t_0) \) in \( B \), we simply set the transmembrane potential to its resting state. We obtain the weak form of the electrical residual (22) through the integration over the domain \( \Omega \), the standard integration by parts, and the inclusion of the Neumann boundary conditions. For the spatial discretization, we discretize the domain of interest \( \Omega \) with \( n_E \) finite elements \( \Omega^e \) as \( \Omega = \bigcup_{e=1}^{n_E} \Omega^e \) and apply the standard isoparametric concept to interpolate the trial functions \( \phi \) and the test functions \( \delta \phi \)

\[
\delta \phi = \sum_{i=1}^{n_E} N^i \delta \phi_i, \quad \phi = \sum_{j=1}^{n_E} N^j \phi_j
\]  

(23)

Here, \( N \) are the standard shape functions on the element level and \( i, j = 1, \ldots, n_E \) are the \( n_E \) element nodes. For the temporal discretization, we suggest a finite difference approximation of the first order time derivative \( \dot{\phi} \)

\[
\dot{\phi} = (\phi - \phi^n) / \Delta t
\]  

(24)

in combination with a standard backward Euler time integration scheme. Using the discretizations in space (23) and time (24), we can introduce the discrete algorithmic residual \( R^n_\phi \)

\[
R^n_\phi = A_{\phi}^{nd} = \int_{\Omega} N^i \dot{\phi} - \frac{\phi^n}{\Delta t} + \nabla N^i \cdot \mathbf{q} \, d\Omega - \int_{\Omega} N^i \mathbf{f} \, d\Omega = 0
\]  

(25)

The operator \( A \) symbolizes the assembly of all element contributions at the element nodes \( i = 1, \ldots, n_E \) to the overall residual at the global node points \( l = 1, \ldots, n_{nd} \). To solve the discrete system of nonlinear equations (25), we suggest an incremental iterative Newton–Raphson solution technique based on the consistent linearization of the residual, which introduces the global iteration matrix \( K^0_{jj} \)

\[
K^0_{jj} = d_{\phi} R^n_\phi = A_{\phi}^{nd} = \int_{\Omega} N^i \frac{1}{\Delta t} N^j + \nabla N^i \cdot \mathbf{D} \cdot \nabla N^j - \mathbf{D} \cdot \nabla f^0 \, d\Omega
\]  

(26)

For each incremental iteration, we update the global vector of unknowns \( \phi_l = \phi_l - \sum_{j=1}^{n_{nd}} K^{-1}_{lj} R^n_j \) at all \( l = 1, \ldots, n_{nd} \) global nodes. At convergence of the local Newton iteration, i.e., at chemical equilibrium, we can evaluate the source term \( f^0(\phi, \mathbf{e}_{ion}, c_{ion}) \) for the electrical problem (25), and its linearization \( d_{j} f^0(\phi, \mathbf{e}_{ion}, c_{ion}) \) for the global Newton iteration (26).

The use of a fully monolithic implicit solution algorithm allows us to apply an adaptive time stepping procedure, for which the time step size is automatically adjusted in response to the number of Newton iterations towards global equilibrium (Wong et al., 2011). In principle, the above equations for \( R^n_\phi \) and \( K^0_{jj} \) are generically portable and can interface with any commercially available finite element package. Here, we embed the proposed algorithm in the general multipurpose nonlinear finite element program FEAP (Taylor, 2011).

7. Computational model of a human heart

To illustrate the features of the proposed algorithm, we model the photostimulation of a human heart stimulated at three different pacing sites. We create a patient-specific heart model from magnetic resonance images taken at different depths along the heart’s long axis, see Fig. 10. On these two-dimensional slices, we use semi–automated image processing tools to section the regions of the cardiac muscle. From the raw and noisy grayscale images, we create monochrome images with sharply defined boundaries using thresholding and binary masking. From the resulting black and white slices, we generate three preliminary triangular surface meshes, one for the outer surface and one for the inner surface of each ventricle. Finally, we convert the surface meshes into a tetrahedral volume mesh consisting of 3129 nodes and 11,347 tetrahedral elements (Kotikanyadanam et al., 2010), see Fig. 10.

We virtually inject our genetically engineered, light sensitive cardiac cells into different regions of the heart (Modi et al., 2011), and model all other cells as conventional cardiac muscle cells (ten Tusscher et al., 2004; Wong et al., 2011). The former group of cells possesses the additional channelrhodopsin current \( I_{ChR2} \), while the latter does not. Following the literature, we model cell injection by modifying the material properties of the myocardial wall at the injection sites (Wenk et al., in press). Here, these regions span a volume of approximately 0.02 cm³, which is roughly the size of the 0.6 × 0.3 × 0.1 cm³ large atrioventricular node in humans, corresponding to approximately 10⁷ cells (James, 1961). To
initiate electrical activation, we photostimulate the light sensitive cells for a period of 30 ms with blue light, then turn off the light, and record the action potential propagation across the heart.

7.1. Atrioventricular node pacing through photostimulation

Fig. 11 illustrates the activation profile in response to atrioventricular node pacing through photostimulation. To reproduce the native activation sequence of the human heart, we virtually inject photosensitive cells into the basal septal region, the location of the atrioventricular node, and pace them with blue light. A depolarization wave forms in the region of the atrioventricular node, travels down the septum, and activates the left and right ventricles. When comparing the temporal evolution of the transmembrane potential $f$, top row, with the different ion concentrations $c_{Na}$, $c_{K}$, and $c_{Ca}$, lower rows, we observe that the sodium concentration changes first, with a sharp front, followed by changes in the potassium and calcium concentrations, with smoother fronts. These observations are in agreement with the temporal evolution of the local ion concentrations illustrated in Fig. 8. After approximately 50 ms, the entire heart is depolarized. The transmembrane potential has changed from approximately $-80$ mV to $+20$ mV.

7.2. Apical pacing through photostimulation

Fig. 12 shows the response of the heart to apical pacing through photostimulation. The right ventricular apex has traditionally been the most prominent pacing site, providing a stable lead position associated with few complications. To predict the activation sequence in response to apical pacing, we virtually inject photosensitive cells into the apex, stimulate them with light for 30 ms, turn off the light, and monitor the electrical and chemical fields. As the transmembrane potential profile indicates, a depolarization wave forms at the apex, travels up towards the base, and activates the septum and both ventricles simultaneously. Again, we observe the rapid increase in the intracellular sodium concentration $c_{Na}$, causing the heart to depolarize. The transmembrane potential changes from $-80$ mV to $+20$ mV within the first 50 ms of the cycle, until the heart is completely depolarized. The intracellular calcium concentration $c_{Ca}$ follows smoothly, while the intracellular potassium concentration $c_{K}$ changes only in the later stages.

7.3. Biventricular pacing through photostimulation

Fig. 13 displays the activation of the heart, photostimulated through bi-ventricular pacing. Biventricular pacing is a common pacing strategy in patients with heart failure for which both ventricles do not contract synchronously. To simulate biventricular pacing, we virtually inject photosensitive cells into the lateral walls of the left and right ventricles and pace both locations simultaneously. In response to photostimulation, two depolarization waves form, one at each pacing site. The waves travel along the ventricles to finally activate the apex and the septum. Again, during depolarization, the transmembrane potential profile $\phi$ closely follows the intracellular sodium concentration $c_{Na}$, which triggers the rapid upstroke of the action potential, while the intracellular calcium concentration $c_{Ca}$ follows slightly later with a smoother propagation front.

8. Discussion

8.1. Comparison with the literature

Within the past decade, optogenetics has become a tremendously active field of research (Deisseroth, 2011), however, little is known about its impact on cardiac cells, neither in isolation nor in the cardiac system. Since the channelrhodopsin photocycle itself is relatively well established from intense studies of the neuronal system (Nikolic et al., 2006, 2009), we simply adopt the kinetics of a well-characterized three-state photocycle model (Hegemann et al., 2005; Nagel et al., 2003).

Creating a cellular model system that expresses Channelrhodopsin-2 allows us to validate its photocurrent using whole cell patch-clamp (Jia et al., 2011) or multielectrode array recordings (Bruegmann et al., 2010). Using both techniques, we have recently demonstrated that our cell model is capable of reliably reproducing experimentally measured photostimulation amplitudes, pulse widths, and frequencies from single cell recordings (Abilez et al., 2011). In this paper, for the first time, we quantify the impact of photostimulation in side-by-side comparisons between conventional cells, electrically stimulated for 1 ms, and genetically engineered cells, optically stimulated for 30 ms, see Figs. 6–9. The gating variables in Fig. 6 can be understood as internal variables, which cannot be measured experimentally. The ionic currents in Fig. 7 can be extracted experimentally from a series of voltage and current clamp recordings using drugs to selectively block the activity of different combinations of channels. Although theoretically possible, for optogenetically engineered cells, typically only the channelrhodopsin photocurrent is measured experimentally. Its indirect impact on all other currents, however, has not been characterized systematically to date. The ion concentrations in Fig. 8 could potentially be measured experimentally, however, for optogenetically engineered cells, this has not been done to date. Unfortunately, only three groups worldwide have reported the use of optogenetics to manipulate the cardiac system, one in zebrafish (Arrenberg et al., 2010), one in transgenic mice (Bruegmann et al., 2010), and one in stem cell derived cardiomyocytes (Abilez et al., 2011). The last study by our own group is the only one that has demonstrated the potential of optogenetics in human heart.

cells. This underlines the importance of computational models, which provide easy access to ion channel dynamics and ionic concentrations, both in healthy and diseased conditions.

The transmembrane potential in Fig. 9 could potentially be extracted from patch clamp or microelectrode array recordings. Here, for the selected light intensity, we find 30 ms to be the minimum light exposure time to evoke a standard action potential. For photostimulation just slightly above this threshold, we identify the time delay between electrical stimulation and photostimulation to be 34 ms, which agrees nicely with the values of 19.7 ± 3.4 ms reported in the literature (Bruegmann et al., 2010). Although not explicitly reported here, in agreement with the literature, we observe a decrease in activation delay for increased light intensities (Bruegmann et al., 2010). Beyond the stimulation threshold, however, electrically and optically activated cells display an almost identical electrophysiology with the characteristic rapid upstroke, early, partial repolarization, a pronounced plateau, a final repolarization, and a return to the resting phase, see Fig. 9. Again, this agrees nicely with experimental findings, which report that channelrhodopsin expression in murine embryonic stem cells is not associated with additional leak currents, or changes in the resting membrane potential, membrane resistance, and action potential duration (Bruegmann et al., 2010).

A first qualitative study in zebrafish has demonstrated that optogenetic tools can, in principle, be used to control the cardiac system in vivo (Arrenberg et al., 2010). In that study, optical perturbation of genetically engineered pacemaker cells was entirely reversible. This agrees nicely with our optically stimulated transmembrane potential returning smoothly to its resting state, see Fig. 9. The study also showed that only few genetically engineered cells are needed to deliver a sufficient optical stimulus in zebrafish hearts (Arrenberg et al., 2010). In our human heart, we virtually inject approximately 10^7 cells covering a volume of 0.02 cm^3, which is roughly the size of the atrioventricular node in humans, see Fig. 10. In agreement with an in vivo study in transgenic mice (Bruegmann et al., 2010), we observe that smaller cell volumes require either increased light intensities or longer stimulation times. Similar to the reported activation delays of 12.2 ± 3.7 ms and 9.3 ± 2.2 ms upon atrial and ventricular photostimulation in mice (Bruegmann et al., 2010), we observe an activation delay of 19.5 ms when comparing our light activated heart in Fig. 11 with previous simulations of native activation sequences (Kotikanyadanam et al., 2010; Tsamis et al., 2011) and electrical pacing (Wong et al., 2011).
Overall, because of the inherent frequency mismatch in rodents and humans, the complete in vivo validation of our model remains challenging, and can only be addressed satisfactorily by means of large animal models. This is, however, a generic limitation of the clinical translational character of cardiac optogenetics in general. Large animal models would also allow us to quantify the impact of optogenetical stimulation on cardiac function in vivo. In a previous study, we have demonstrated how light can be turned into force using optogenetic tools in vitro (Abilez et al., 2011). A related study by another group reported light-induced mechanical strains of up to 4.5% (Jia et al., 2011). We are currently integrating our continuum model for the photoelectrochemistry of living systems into a model for excitation–contraction coupling (Göktepe et al., 2010). This will allow us to characterize the mechanically-mediated ionic response of strain-gated ion channels (Reeves et al., 2008; Ursell et al., 2007), and, vice versa, the calcium-mediated mechanical response of the heart (Criscone et al., 2002; Rausch et al., 2011), and its impact on muscle contraction and cardiac function (Givli and Bhattacharya, 2009; Göktepe et al., 2010).

8.2. Conclusion

At an unprecedented temporal and spatial precision, optogenetic tools enable us to manipulate electrically active cells with light. This study documents our first attempt to model genetically engineered light-sensitive heart muscle cells, with the ultimate goal to control the cardiac system by mean of photostimulation. In contrast to electrical pacemakers, light delivery is minimally invasive, genetically targeted, and temporally precise. Most importantly, light can be delivered at a distance. Unlike pacing leads for electrical stimulation, which are known to have a high perforation risk, the light source for optical stimulation does not need to be in direct contact with the moving heart muscle. Light pacing might therefore be an attractive remote, less invasive, and more durable alternative to conventional electrical pacing. Unlike traditional electrical stimulation, optogenetic stimulation allows us to precisely control the selective permeability of the plasma...
membrane, its conductivity with respect to different ions, its sensitivity to light of different wavelengths, and the spatio-temporal evolution of different opening and closing profiles. Given this incredible freedom, there is a pressing need to establish economic strategies to optimize the matrix of input variables. Predictive continuum models for photoelectrochemistry allow us to virtually probe landscapes of process parameters and identify optimal photostimulation sequences in various different tissues and organs. Discretized with finite element tools, they enable the combination of virtually any cell type on virtually any geometry. Our time-adaptive algorithm is highly efficient, conceptually modular, and easily portable to any commercial nonlinear finite element platform. While the photoelectrochemical behavior of living tissue is almost impossible to characterize in vivo, and little is known about the spatio-temporal distribution of sodium, potassium, and calcium ions, computational tools like ours allow us to predict action potential profiles and ionic concentrations across the heart in silico within the order of minutes. We believe that computational optogenetics will be widely applicable to predict the response of other genetically engineered, electrically-active cells and, ultimately, support the design of novel therapies for various types of neuronal, musculoskeletal, pancreatic, and cardiovascular disorders such as depression, schizophrenia, cerebral palsy, paralysis, diabetes, pain syndromes, and cardiac arrhythmias.

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Appendix A

Our cardiac cell model is based on a well-characterized and frequently used ventricular cell model (ten Tusscher et al., 2004; Wong et al., 2011), enhanced by the channelrhodopsin photocurrent $I_{\text{ChR2}}$ and its corresponding gating variable $g_{\text{ChR2}}$. We characterize the cell through $n_{\text{ion}} = 4$ ion concentrations

$$c_{\text{ion}} = [c_{\text{Na}}, c_{\text{K}}, c_{\text{Ca}}, c_{\text{Ca}}^\text{in}]$$

where $c_{\text{Na}}$, $c_{\text{K}}$, and $c_{\text{Ca}}$ are the intracellular sodium, potassium, and calcium concentrations, and $c_{\text{Ca}}^\text{in}$ is the calcium concentration in the sarcoplasmic reticulum. Changes in these concentrations are brought about by an in- and outflux of ions which we describe through a total of $n_{\text{g}} = 16$ ionic currents

$$I_{\text{g}} = [I_{\text{Na}}, I_{\text{K}}, I_{\text{NaK}}, I_{\text{NaCa}}, I_{\text{ChR2}}, I_{\text{Kr}}, I_{\text{pCa}}, I_{\text{fCa}}, I_{\text{up}}, I_{\text{ref}}]$$

These are the fast sodium current $I_{\text{Na}}$, the background sodium current $I_{\text{bNa}}$, the sodium potassium pump current $I_{\text{NaK}}$, the sodium calcium exchanger current $I_{\text{NaCa}}$, the channelrhodopsin photocurrent $I_{\text{ChR2}}$, the inward rectifier current $I_{\text{Kr}}$, the rapid delayed rectifier current $I_{\text{Kr}}$, the slow delayed rectifier current $I_{\text{Kr}}$, the plateau potassium current $I_{\text{pK}}$, the transient outward current $I_{\text{to}}$, the L-type calcium current $I_{\text{CaL}}$, the background calcium current $I_{\text{CaB}}$, the plateau calcium current $I_{\text{pCa}}$, the leakage current $I_{\text{fCa}}$, and the sarcoplasmic reticulum uptake and release currents $I_{\text{up}}$ and $I_{\text{ref}}$. The activation and inactivation of these currents is governed by the transmembrane potential $\phi$, and by $n_{\text{g}} = 14$ gating variables

$$g_{\text{g}} = [g_{\text{m}}, g_{\text{h}}, g_{\text{f}}, g_{\text{ChR2}}, g_{\text{Kr}}, g_{\text{pK}}, g_{\text{Ca}}, g_{\text{fCa}}]$$

In particular, these are the sodium activation gate $g_{\text{m}}$, the fast and slow sodium inactivation gates $g_{\text{h}}$ and $g_{\text{f}}$, the channelrhodopsin activation gate $g_{\text{ChR2}}$, the rapid delayed rectifier activation and inactivation gates $g_{\text{Kr}}$, and $g_{\text{Kr}}$, the slow delayed rectifier activation gate $g_{\text{fCa}}$, the transient potassium activation and inactivation gates $g_{\text{pK}}$, and $g_{\text{pK}}$, and the calcium release activation gate $g_{\text{fCa}}$. We will now specify the mathematic model for the concentrations $c_{\text{ion}}$, currents $I_{\text{g}}$, and gating variables $g_{\text{g}}$. In Table 1, we summarize all model parameters, their physical interpretations, and their parameter values (ten Tusscher et al., 2004; Wong et al., 2011).

A.1. Sodium concentration, currents, and gating variables

Sodium plays a crucial role in generating the fast upstroke in the initial phase of the action potential. At rest, both electrical forces and chemical gradients pull extracellular sodium ions into the cell. The influx of sodium ions is small, however, since at rest, the membrane is relatively impermeable to sodium. An external stimulus above a critical threshold value, here generated through either photostimulation or through active neighboring cells, opens the fast sodium channels to initiate a rapid inflow of sodium ions associated with the rapid depolarization of the cell membrane. The transmembrane potential increases drastically by more than 100 mV in less than 2 ms, see Fig. 9. At the end of the upstroke, the cell membrane is positively charged, and the fast sodium channels return to their closed state. The sodium concentration

$$\dot{c}_{\text{Na}} = -\frac{C}{V} [I_{\text{Na}} + I_{\text{bNa}} + 3I_{\text{NaK}} + 3I_{\text{NaCa}} + I_{\text{ChR2}}]$$

The sodium related currents are defined as follows: three indicates that both the sodium potassium pump and the sodium calcium exchanger operate at a three-to-two ratio.

For changes in response to the fast sodium current \( I_{Na} \), the background sodium current \( I_{bNa} \), the sodium potassium pump current \( I_{NaK} \), and the sodium calcium exchanger current \( I_{NaCa} \), scaled by the membrane capacitance per unit surface area \( C = 185 \text{ pF} \), the cytoplasmic volume \( V = 16.404 \text{ \mu m}^3 \), and the Faraday constant \( F = 96.4867 \text{ C/mmol} \). The scaling factor three indicates that both the sodium potassium pump and the sodium calcium exchanger operate at a three-to-two ratio. The sodium related currents are defined as follows:

\[
\begin{align*}
I_{Na} &= C_{Na}^{\max} \delta \left( \phi - \phi_{Na} \right) \\
I_{bNa} &= C_{bNa} \left( \phi - \phi_{bNa} \right) \\
I_{NaK} &= I_{NaK}^{\max} \left[ C_{NaK} + C_{bNaK} \right] \\
I_{NaCa} &= I_{NaCa}^{\max} \left[ C_{NaCa} + C_{CaNa} \right] \\
I_{CaR} &= C_{CaR}^{\max} \delta \left( \phi - \phi_{CaR} \right)
\end{align*}
\]
The sodium potassium pump inactivation of the sodium channel almost instantaneously after the rapid upstroke, we express the steady state value and cytoplasmic volume role. The six potassium related currents scale by the membrane capacitance per unit surface area mainly regulated by the rapid and slow delayed rectifier currents observe an influx of calcium ions which is balanced by the efflux of an equal amount of positively charged potassium ions, transient outward current repolarization governed by the voltage-activated transient outward current.

The kinetics of inactivation are exponential. For the fast sodium inactivation gate, $g_m = [g_m^{∞} - g_m]/τ_m$, which initiates the rapid upstroke, they take the following explicit representations:

$$
\begin{align*}
\tau_m &= 0.1(1 + e^{-(60-ϕ)/5})^{-1} + [1 + e^{(ϕ+35)/5}]^{-1} + [1 + e^{(ϕ-50)/200}]^{-1} \\
g_m^{∞} &= 1 + e^{(ϕ+71.55)/7.43}^{-2}
\end{align*}
$$

The sodium potassium pump $I_{NaK}$, a metabolic pump that continuously expels sodium ions from the cell interior and pumps in potassium ions, is responsible to remove the sodium ions, which had entered the cell rapidly during the fast upstroke. The sodium calcium exchanger $I_{NaCa}$ also affects the intracellular sodium concentration through the expulsion of intracellular calcium ions. The additional parameters that characterize the sodium potassium pump current $I_{NaK}$ and for the sodium calcium exchanger current $I_{NaCa}$ are the extracellular sodium, potassium, and calcium concentrations $c_{Na}$, $c_{K}$, and $c_{Ca}$, the half saturation constants $c_{Na}^{max}$, $c_{NaCa}^{max}$, $c_{NaK}^{max}$, and $c_{Ca}$, the half saturation factors $k_{NaCa}^{sat}$, and the voltage dependent sodium calcium parameter $γ_{NaCa} = 1.2$.

### A.2. Potassium concentration, currents, and gating variables

Potassium plays an important role in maintaining the appropriate action potential profile in all four phases after the rapid upstroke. Unlike for sodium, the electrical force that pulls potassium ions inward is slightly weaker than the chemical force of diffusion pulling potassium ions outward. Accordingly, potassium tends to leave the cell, but not at the same rate as sodium. The sodium potassium pump current $I_{NaK}$ and the plateau potassium current $I_{K}$ play a minor role. The six potassium related currents scale by the membrane capacitance per unit surface area $C=185 \text{ pF}$, by the cytoplasmic volume $V = 16.404 \mu\text{m}^3$, and by the Faraday constant $F=96.487 \text{ C/mmol}$. We adopt their following

$$
\begin{align*}
c_K &= -\frac{C}{VF}[I_{K1} + I_{K2} + I_{K3} - 2I_{NaK} + I_{K4} + I_{K0}]
\end{align*}
$$

subsection, we have introduced the extracellular sodium and potassium concentrations and therefore characterized differently in different regions of the heart. Similar to the previous transient potassium outward current and slow delayed rectifier current $I_{Kr}$ and $I_{Ks}$ govern the potassium efflux. The channel for the rapid delayed rectifier current $I_{Kr}$ is gated by an activation gate $g_{x1} = [g_{x1}^N - g_{x1}]/\tau_{x1}$ with the steady state value and time constant given as:

$$g_{x1} = [1 + e^{-(26 - \phi)}/7]^{-1}$$  \(\quad \text{(38)}\)

and by an inactivation gate $g_{x2} = [g_{x2}^N - g_{x2}]/\tau_{x2}$, with the following steady state value and time constant:

$$g_{x2} = [1 + e^{(6 + 88)/24}]^{-1}$$  \(\quad \text{(39)}\)

The channel for the slow delayed rectifier current $I_{Ks}$ is a function of the reversal potential $\phi_{Ks} = RT/F \log([c_{k0} + P_{KNa}c_{Na}]/[c_{K} + P_{KNa}c_{Na}])$ parameterized in terms of its permeability to sodium ions $P_{KNa} = 0.03$. It is gated by an activation gate $g_{s} = [g_{s}^N - g_{s}]/\tau_{s}$ in terms of the following parameterization:

$$g_{s} = [1 + e^{-(5 - \phi)/14}]^{-1}$$  \(\quad \text{(40)}\)

The transient potassium outward current $I_{0}$ is responsible for the transition between the rapid upstroke and the plateau phase, where it generates an early short period of limited repolarization. It is gated by a voltage-dependent activation gate $g_{r}$ with $g_{r} = [g_{r}^N - g_{r}]/\tau_{r}$ defined through the following steady state value and time constant:

$$g_{r} = [1 + e^{(20 - \phi)/6}]^{-1}$$  \(\quad \text{(41)}\)

and by the voltage-dependent inactivation gate $g_{s}$ with $g_{s} = [g_{s}^N - g_{s}]/\tau_{s}$ with the steady state value and time constant given as follows:

$$g_{s} = [1 + e^{(5 + 20)/5}]$$  \(\quad \text{epicardium,} \quad g_{s} = [1 + e^{(5 + 28)/5}]\)

$$\tau_{s} = 85e^{-(\phi + 45)/320} + 5[1 + e^{(\phi - 20)/5}] + 3 [\text{endocardium,} \quad \tau_{s} = 1000e^{-(\phi + 67)/1000 + 8}] \quad \text{(42)}\)

This voltage dependent potassium inactivation gate displays a significantly different behavior for epicardial and endocardial cells and is therefore characterized differently in different regions of the heart. Similar to the previous subsection, we have introduced the extracellular sodium and potassium concentrations $c_{Na0} = 140$ mM and $c_{K0} = 5.4$ mM, and the half saturation constants $c_{KNa} = 1$ mM and $c_{NaK} = 40$ mM.
A.3. Calcium concentration, currents, and gating variables

Calcium is the key player to translate electrical excitation into mechanical contraction. During the plateau of the action potential, calcium ions enter the cell through calcium channels that typically activate and inactivate smoothly and more slowly than the fast sodium channels. The influx of positively charged calcium ions through the L-type calcium channel $I_{\text{Ca}}$ is balanced by an efflux of positively charged potassium ions. The letter $L$ indicates the long lasting nature of the inward calcium current. Overall, the intracellular calcium concentration evolves in response to seven currents

$$\dot{c}_{\text{Ca}} = \frac{C}{2VF}[I_{\text{Ca}} + I_{\text{Ca}} + I_{\text{pCa}} - 2I_{\text{NaCa}}] + I_{\text{leak}} - I_{\text{up}} + I_{\text{rel}}]$$ (43)

the L-type calcium current $I_{\text{Ca}}$, the background calcium current $I_{\text{Ca}}$, the plateau calcium current $I_{\text{pCa}}$, and the sodium calcium pump current $I_{\text{NaCa}}$, weighted by the membrane capacitance per unit surface area $C=185\, \mu\text{F/cm}^2$, the cytoplasmic volume $V=16,404\, \mu\text{m}^3$, and the Faraday constant $F=96.487\, \text{C/mol}$. In addition, the leakage current $I_{\text{leak}}$, the sarcoplastic reticulum uptake current $I_{\text{up}}$, and the sarcoplastic reticulum release current $I_{\text{rel}}$ affect the intracellular calcium concentration through a calcium exchange with the sarcoplastic reticulum. We adopt the following definitions for the individual calcium related currents:

$$I_{\text{Ca}} = C_{\text{Ca}}^\text{max} g_{\text{Ca}} f_{\text{Ca}} [4\phi^2]/[RT] [c_{\text{Ca}}^2/2F/\text{RT}] - 0.341 c_{\text{Ca}} [e^{2\phi}/\text{RT} - 1]^{-1}$$

where the individual scaling factors are the maximum calcium conductance $C_{\text{Ca}}^\text{max} = 0.175\, \text{mm}^3\, \mu\text{F}^{-1}\, \text{s}^{-1}$, the maximum background calcium conductance $C_{\text{Ca}}^\text{max} = 0.000592\, \text{nS}/\text{pF}$, the maximum plateau calcium conductance $C_{\text{Ca}}^\text{max} = 0.825\, \text{nS}/\text{pF}$, the maximum sodium calcium pump current $I_{\text{NaCa}}^\text{max} = 1000\, \text{pA}/\text{pF}$, the maximum leakage current $I_{\text{leak}}^\text{max} = 0.08\, \text{s}^{-1}$, the maximum sarcoplasmatic reticulum calcium uptake current $I_{\text{up}}^\text{max} = 0.000425\, \text{mM/s}$, and the maximum sarcoplasmatic reticulum calcium release current $I_{\text{rel}}^\text{max} = 8.232\, \text{mM/s}$. The major calcium channel, the long-lasting L-type calcium channel $I_{\text{Ca}}$, opens and closes in response to the voltage-dependent activation gate $g_{\text{a}} = [g_{\text{a}}^\infty - g_{\text{a}}]/\tau_{\text{a}}$ characterized through the following steady state value and time constant

$$g_{\text{a}}^\infty = [1 + e^{-5/(\phi/7.5)}]^{-1}$$

in response to the voltage-dependent inactivate gate $g_{\text{f}} = [g_{\text{f}}^\infty - g_{\text{f}}]/\tau_{\text{f}}$ characterized through

$$g_{\text{f}}^\infty = [1 + e^{(\phi + 20)/7}]^{-1}$$

and in response to the intracellular calcium dependent inactivation gate $g_{\text{Ca}} = [g_{\text{Ca}}^\infty - g_{\text{Ca}}]/\tau_{\text{Ca}}$ characterized through

$$g_{\text{Ca}}^\infty = 0.685(1 + (e^{(\phi - 0.000325)}/0.000325)^8)^{-1} + 0.1(1 + e^{(\phi - 0.00055)/0.0001})^{-1} + 0.2(1 + e^{(\phi - 0.00075)/0.0008})^{-1} + 0.23$$

$$\tau_{\text{Ca}} = \begin{cases} \infty & \text{if } g_{\text{Ca}}^\infty > g_{\text{Ca}} \text{ and } \phi \geq -60\, \text{mV} \\ 2\, \text{ms} & \text{otherwise.} \end{cases}$$ (47)

Accordingly, the steady state response $g_{\text{Ca}}^\infty$ has a switch-like shape when going from no inactivation to considerable but incomplete inactivation, depending mildly on the calcium concentration $C_{\text{Ca}}$ for suprathreshold concentrations. Last, the calcium-induced calcium release current $I_{\text{rel}}$ is characterized through the activation gate $g_{\text{a}}$, the same gate that is also activating the L-type calcium channel of $I_{\text{Ca}}$, and through the calcium-dependent inactivation gate $g_{\text{Ca}} = [g_{\text{Ca}}^\infty - g_{\text{Ca}}]/\tau_{\text{Ca}}$ characterized through the following steady state value and time constant:

$$g_{\text{Ca}}^\infty = \begin{cases} [1 + e^{\phi}/0.000356]^8 & \text{if } C_{\text{Ca}} \leq 0.00035 \\ [1 + e^{(C_{\text{Ca}} - 0.00035)/0.003}]^{-1} & \text{otherwise.} \end{cases}$$

The remaining parameters governing the response of the plateau calcium current $I_{\text{pCa}}$, the calcium uptake current $I_{\text{up}}$, and the sarcoplastic reticulum calcium release current $I_{\text{rel}}$ are the half saturation constants for the plateau calcium concentration $c_{\text{Ca}}^p = 0.0005$ mM, for the sarcoplastic reticulum calcium uptake current $c_{\text{up}} = 0.00025$ mM, and for the sarcoplastic reticulum calcium release $c_{\text{rel}} = 0.25$ mM, respectively. The parameter $\gamma_{\text{NaCa}} = 2.5$ enhances the outward nature of the sodium calcium pump current $I_{\text{NaCa}}$. The additional parameter $\gamma_{\text{el}} = 2$ weighs the relative influence of the sarcoplastic reticulum calcium concentration on sarcoplastic reticulum calcium release $I_{\text{rel}}$. Finally, we represent the total intracellular calcium concentration $c_{\text{Ca}}^\text{tot} = c_{\text{Ca}} + c_{\text{Ca}}^\text{buf}$ in the cytoplasm as the sum of the free intracellular calcium concentration $c_{\text{Ca}}$ and the buffered calcium concentration $c_{\text{Ca}}^\text{buf} = [c_{\text{Ca}}c_{\text{buf}}^\text{Ca}][c_{\text{Ca}} - c_{\text{Ca}}^\text{buf}]^{-1}$. Accordingly, we scale the definition of the free intracellular calcium concentration in Eq. (43) with the parameter $\gamma_{\text{Ca}} = [1 + [c_{\text{Ca}}c_{\text{buf}}][c_{\text{Ca}} + c_{\text{buf}}^{-1}]]^{-1}$, where $c_{\text{Ca}} = 0.15$ mM and $c_{\text{buf}} = 0.001$ mM are the total and half saturation cytoplasmic calcium buffer concentrations.

### A.4. Sarcoplastic reticulum calcium concentration, currents, and gating variables

The specification of the sarcoplastic reticulum calcium concentration

$$c_{\text{Ca}}^\text{sr} = \frac{V}{V_{\text{rel}}} [-I_{\text{leak}} + I_{\text{up}} - I_{\text{rel}}]$$  \hspace{1cm} (49)$$

is now relatively straightforward since it mimics the corresponding loss of intracellular calcium scaled by the ratio between the volume of the cytoplasm $V = 16,404$ μm$^3$ and the volume of the sarcoplastic reticulum $V_{\text{rel}} = 1094$ μm$^3$. The leakage current $I_{\text{leak}}$, the sarcoplastic reticulum uptake current $I_{\text{up}}$, and the sarcoplastic reticulum release current $I_{\text{rel}}$ are defined as before

$$I_{\text{leak}} = I_{\text{leak}}^\text{max}[c_{\text{Ca}} - c_{\text{Ca}}]$$

$$I_{\text{up}} = I_{\text{up}}^\text{max}[1 + c_{\text{Ca}}^2/c_{\text{Ca}}^2]^{-1}$$

$$I_{\text{rel}} = I_{\text{rel}}^\text{max} g_{\text{Ca}} g_{\text{Ca}}^2 [1 + \gamma_{\text{el}} c_{\text{Ca}}^2 [c_{\text{Ca}} + c_{\text{Ca}}^2/2]^{-1}]$$  \hspace{1cm} (50)$$

We have already introduced the maximum leakage current $I_{\text{leak}}^\text{max} = 0.08$ s$^{-1}$, the maximum sarcoplastic reticulum calcium uptake current $I_{\text{up}}^\text{max} = 0.000425$ mM/s, and the maximum sarcoplastic reticulum calcium release current $I_{\text{rel}}^\text{max} = 8.232$ mM/s, the half saturation constants for the calcium uptake $c_{\text{up}} = 0.00025$ mM, and for the calcium release $c_{\text{rel}} = 0.25$ mM, and the weighting coefficient $\gamma_{\text{Ca}} = 2$ in the previous subsection. Finally, we introduce the total calcium concentration in the sarcoplastic reticulum $c_{\text{Ca}}^\text{sr} = c_{\text{Ca}} + c_{\text{Ca}}^\text{buf}$ as the sum of the free sarcoplastic reticulum calcium concentration $c_{\text{Ca}}^\text{sr}$ and the buffered sarcoplastic reticulum calcium concentration $c_{\text{Ca}}^\text{buf} = [c_{\text{Ca}}c_{\text{buf}}][c_{\text{Ca}} - c_{\text{Ca}}^\text{buf}]^{-1}$. Accordingly, we scale the free sarcoplastic reticulum calcium concentration in Eq. (49) by the parameter $\gamma_{\text{Ca}}^\text{sr} = [1 + [c_{\text{Ca}}c_{\text{buf}}][c_{\text{Ca}} + c_{\text{buf}}^{-1}]]^{-1}$, where $c_{\text{Ca}}^\text{tot} = 10$ mM and $c_{\text{buf}} = 0.3$ mM are the total and half saturation sarcoplastic reticulum calcium buffer concentrations.

### References


