Implementation of an electrophysiological model of human-induced pluripotent stem cell-derived cardiomyocytes

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My working environment

Barcelona Supercomputing Center (BSC) is among the largest centers for specialization in high performance computing. As a master student in the field of computational mechanics interested to gain knowledge about the most up-to-date methods and tools for better computational advancements in the current world of research and technology, this center attracted my attention from the day I was given the chance to visit the whole supercomputing department and be explained about the job done there.

As a student involved in computational projects, I can appreciate the fast and optimal method used at BSC to perform research along with the powerful computers available, which fired my enthusiasm to seek a project there. Hopefully, I got the opportunity to attend one of the lectures held at UPC given by Mariano Vazquez who is one of the best researchers at BSC. I became motivated in his field of research, cardiovascular system simulations, and had the the chance to participate in one of his group projects. I was honored to work under supervision of Jazmin Aguado Sierra who is also one of the best researchers working in the field of biomechanics.

Working with her on the topic of this internship was one of my best experiences in doing research. I highly appreciate her collaboration, her knowledge and her guidance whenever we faced an issue. I believe my working environment at BSC provided a context in which I could improve my knowledge and achieve the most satisfactory outcomes from my job. As another critical factor of my working environment, I can mention the eagerness of the general atmosphere to investigate, learn and move on the cutting edge of the science. This helped me become even more motivated about the fascinating aspects of knowledge waiting above the university level of education to be discovered. In addition, this gave me the opportunity to not only learn interesting up-to-date scientific materials but also put my knowledge, which I gained from university, into practice.

This, without any doubt, shed light into the dark corners of my knowledge and helped me to assimilate them deeply and integrate them all and make a consolidated knowledge basis which will definitely help me in my future career. All in all, I consider my working experience at BSC as a breakthrough for myself in my field of research.

Abstract

The fields of medicine and physiology have gone through an enormous increase in the utilization of computer simulations to enhance and understand the basic biology and pathology which are under investigation in laboratories. The purpose to integrate the biological aspect of the model with mathematical formulations is to deepen the understanding about normal and diseased states to translate basic biology knowledge into clinical applications. In this work, an in-vitro model of human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CM) is implemented with reference to a computational model in literature and analyzed for future inclusion into a finite element code developed at the Barcelona Supercomputing Center called Alya. From the mathematical point of view, the hiPSC-CM model consists of 18 ordinary differential equations which hold for two different cell types namely atrial-like and ventricular-like phenotype through variating some parameters and equations of the whole model. To achieve the numerical solution of this system of equations, the choice of explicit Runge-Kutta fourth order is offered remarking the fact that electrophysiological ion channel models are generally considered as stiff systems given their fast dynamics. The models are successfully implemented within MATLAB and FORTRAN to raise the possibility to reproduce cardiac safety laboratory tests using mathematical models. The main aim of this work is to expand the use of mathematical modeling for advancement of the diagnostic and treatment of the cardiac arrhythmia.

1 Introduction

Recent studies have been focused on deriving the pluripotent stem cells from human tissues, which is considered as a breakthrough upon health providers. The application of pluripotent stem cells is appreciated in basic biology, drug development, and transplantation, [1-3].

Many studies have been carried out to develop reliable models to simulate the electrophysiology of pluripotent stem cells and investigate the in-vitro and in-vivo functionality of them. Most of the developments in this regard are achieved by experimental investigations along with numerical tools to obtain the desired model characterization, [4-6].

The use of mathematical and computational tools to study the human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CM) provides the context to obtain a deeper understanding of cardiomyocytes (CM) function. The significance of this study is appreciated in the perspective of restoring the functionality and repairing infarcted cardiac areas as well as its promising role in the in-vitro drug models.

In order to improve the comprehension of hiPSC cardiomyocytes, an outstanding mathematical model accessible in the literature is taken into consideration, [7]. The mathematical model has been developed based on the experimental data published in [6] discriminating among two different records of action potentials namely ventricular-like and atrial-like hiPSC-CMs.

The experimental data in [6] suggests the gating properties of seven ionic currents including sodium (I_{Na}), L-type calcium (I_{CaL}), hyperpolarization-activated pacemaker (I_f), transient outward potassium (I_{to}), inward rectifier potassium (I_{K1}), and the rapidly and slowly activating components of delayed rectifier potassium (I_{Kr} and I_{Ks}, respectively) current. This provides the possibility of deriving the mathematical formulation for the membrane potential and the aforementioned ionic currents to study the hiPSC-CM from a mathematical viewpoint.

As mentioned before, the analysis of pluripotent stem cells provides the possibility to analyze the responsiveness of hiPSC-CM to various drugs. This advancement is highly crucial in the sense that, as it is an in-vitro model, it clarifies a patient's drug responsiveness before the drug is directly injected to the body; therefore, it is considered as a new platform in the field of

pre-clinical cardiotoxicity and pro-arrhythmia screening of drugs in development, [8].

The aim of this essay is to implement the computational model of hiPSC-CM by means of numerical methods. The model is being prepared to be implemented into a FEM code, Alya [9], to be able to reproduce monolayer experimental data published in [8].

2 Material and method

In order to define the equations describing the model, a schematic diagram of the model depicting cell compartments and major functional components is shown in Figure1, which accounts for both ventricular-like and atrial-like models. The ion currents, namely I_{K1}, I_{to}, I_{Kr}, I_{Ks}, I_{CaL}, I_{NaK}, I_{Na}, I_{NaCa}, I_{pCa}, I_f, I_{bNa}, I_{bCa}, and the membrane potential obey the so-called Hodgkin-Huxley formulation suggesting that the concepts of electronics can be taken advantage of in order to characterize the relations and formulation of the cardiac physiological models [10]. These models can be treated mathematically as electrical circuits describing the relation between currents and voltage of the model as following:

$$C\frac{dV}{dt} = -I_{ion} = -(I_{K1} + I_{to} + I_{Kr} + I_{Ks} + I_{CaL} + I_{NaK} + I_{Na} + I_{NaCa} + I_{pCa} + I_f + I_{bNa} + I_{bCa})$$

where C is cell capacitance and V the membrane voltage.

The formulation of currents were derived by Paci et al.[7], using the experimental data recorded by Ma et al.[6]. The ion currents and consequently the membrane potential, which are of interest in this study, are functions of the membrane potential, V, the reversal potentials, E_{Na}, E_K, E_{Ks}, E_{Ca}, E_f, and the gate parameters related to the currents of intracellular and extracellular ionic concentrations, namely Na^+, Ca^{2+} and K^+. All model equations and parameter values are provided in the Appendix.

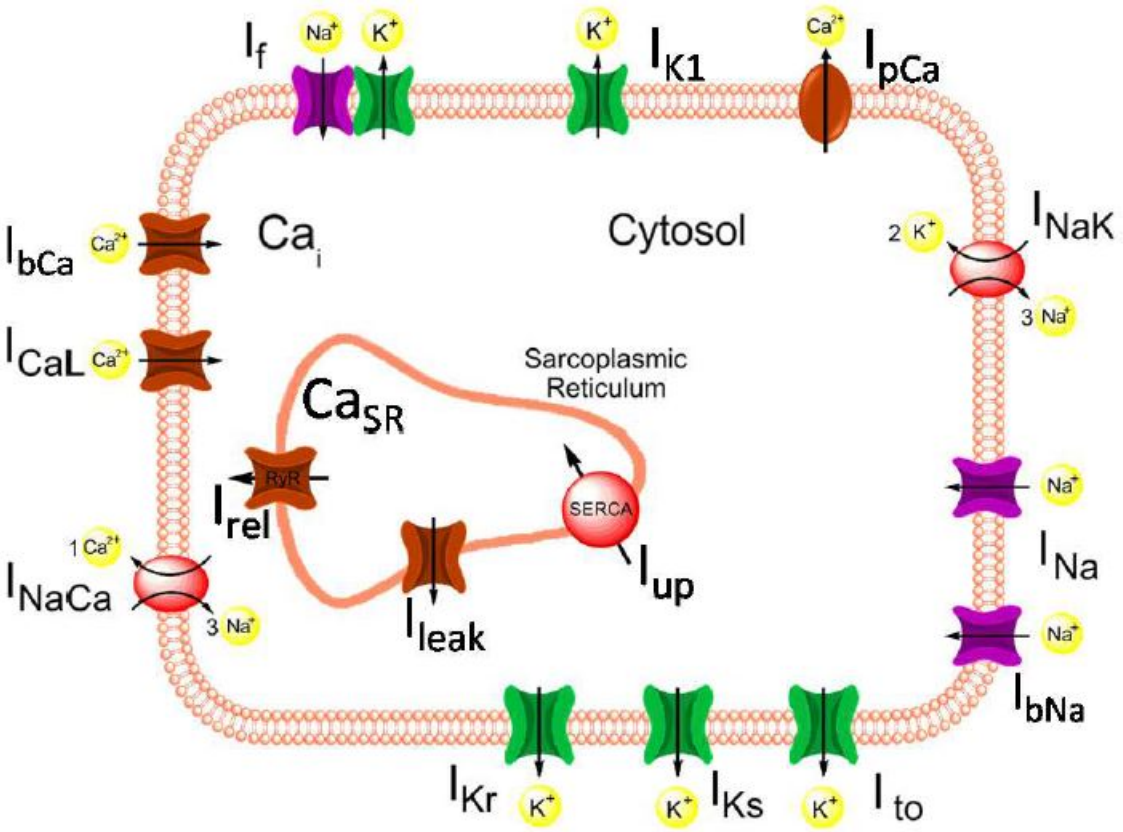


Figure 1: Schematic diagram of the model.

In order to implement the model, all the equations should be defined precisely in the desired coding language. Since the current model is a biological model and the results obtained from the study are going to be used in the future investigations on human issues, this step should be given careful consideration due to the complexity of the equations.

The model generally consists of 18 ordinary differential equations which are coupled. This system of equations should be solved using numerical methods such as the Euler method, central difference or Runge-Kutta method. Considering the fact that electrophysiological cardiac models are stiff initial value problems (IVP), there are different methods which can be appreciated to solve the system. In this study, the explicit method of Runge-Kutta fourth order (RK4) has been chosen since it provides good precision and the convergence of the error to the analytical solution of the system is generally considered to be satisfactory.

Explicit Runge-Kutta fourth order method is considered as a four-stage method. This method is among the most popular ones as it is explicit and the solution of each time step can be computed successively from the previous time step. In addition, this method as it is simple, it provides great precision and decreases the computational costs for complex problems with huge equations as in the current case. The formulation of this iterative method is found in the following:

$$Y_{i+1} = Y_i + \tfrac{1}{6}(K_1 + 2K_2 + 2K_3 + K_4)$$

where

$$\begin{cases} K_1 = f(Xi, Yi) \\ K_2 = f(X_i + \frac{h}{2}, Y_i + \frac{h}{2}K_1) \\ K_3 = f(X_i + \frac{h}{2}, Y_i + \frac{h}{2}K_2) \\ K_4 = f(X_i + h, Y_i + hK_3) \end{cases}$$

and f_i describes the formulation of each ordinary differential equation.

A stiff IVP refers to a system in which the time step size should be chosen based on the stability criteria depending on the method used to solve the problem. In such systems, the step size chosen provides much more accuracy than what is actually essential. This is because of the reason that in stiff problems stability takes on more importance than accuracy, [11].

In order to find the stability region for a specific problem, different studies are done and methods are proposed based on the desired iterative method. For the current complex system of ordinary differential equations being solved using the method of RK4, finding the stability region requires high mathematical analysis which is out of the bounds of this study. However, the possibility still exists for more simple cases as suggested by [12]. Consequently, in the present study, an interval of time steps is defined and this one-dimensional space is searched randomly to find the approximate interval in which the stable solution lies.

After finding the stability region and the interval of the time step by which the solution is assumed to be stable, the accuracy of the solution should be investigated. One of the popular methods in this regard is to compare the solution obtained from different time steps with a reference solution on which the problem can rely. This issue becomes complicated when there is no exact solution to the system. In this case, results can be compared to the solution from a high-order, variable step implicit solver such as the solvers provided by MATLAB.

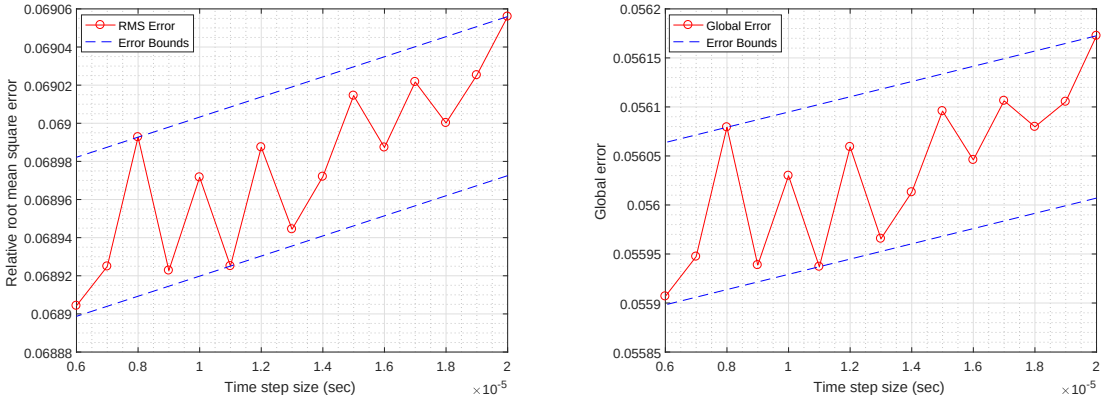
The current electrophysiological model is solved using explicit RK4 with different time steps all of which lie in the stability region and the results are then compared to the solution obtained by [7] which used the MATLAB command, ode15s, to solve this system of equations. This comparison is made through defining a relative root mean squared (RRMS) error for the value of the membrane potential for ventricular-like phenotype. Defining this kind of error is considered very popular and widely acceptable in literature for cardiac simulations [13]. The formula of RRMS error is provided as the following:

$$RRMS = \sqrt{\frac{\sum_{i=1}^{n}(V - V_{Ref})^2}{\sum_{i=1}^{n} V_{Ref}^2}}$$

In addition to calculating the RRMS error, a global error is defined by of the maximum difference of the reference solution and the solution obtained by explicit RK4 method as the following:

$$E_{global} = max(abs(V_{Ref} - V))$$

The error is calculated for 20 values of time step size in the interval defined as $[0.6 \cdot 10^{-5}, 2 \cdot 10^{-5}]$ seconds where the solution to the problem is assumed to be stable. Taking the membrane potential of ventricular-like cell type into account, the graph showing the values of two types of errors previously defined is shown in Figure2.



(a) Relative root mean square error

(b) Global error

Figure 2: Value of the errors between the reference solution and the solution of explicit RK4

As it can be observed, in both figures depicting the RRMS error, Figure2a, and the global error, Figure2b, the range between which the error varies can be considered negligible. In order to get a better insight, an error bound is defined based on the local maximum and minimums of the curves. The width of this error bound for both types of errors is shown in Table1.

Table 1: Error bounds of membrane potential for ventricular-like phenotype

Error type	RRMS	Global
Width of error bound	8.3437e-05	1.6554e-04

The global and the RRMS errors are of order 4 and 5, respectively, as suggested by the table, which means that the given time step size interval is acceptable. In addition, observing the values of based on the provided figures, the errors suggest 5 to 6% which is a reasonable value based on the experimental error within the biology laboratories and the data provided by literature for cardiac simulations.

After implementation of the numerical algorithm and studying the errors, the simulation is performed in 5 seconds with a step size of 10^{-5} both in MATLAB and FORTRAN. The ventricular-like action potential is compared to the reference solution and shown in Figure3. As it can be seen, the results are in great agreement with the reference solution which means the value of step size chosen for this particular method is reliable for future investigations of ion currents and potentials for both phenotypes.

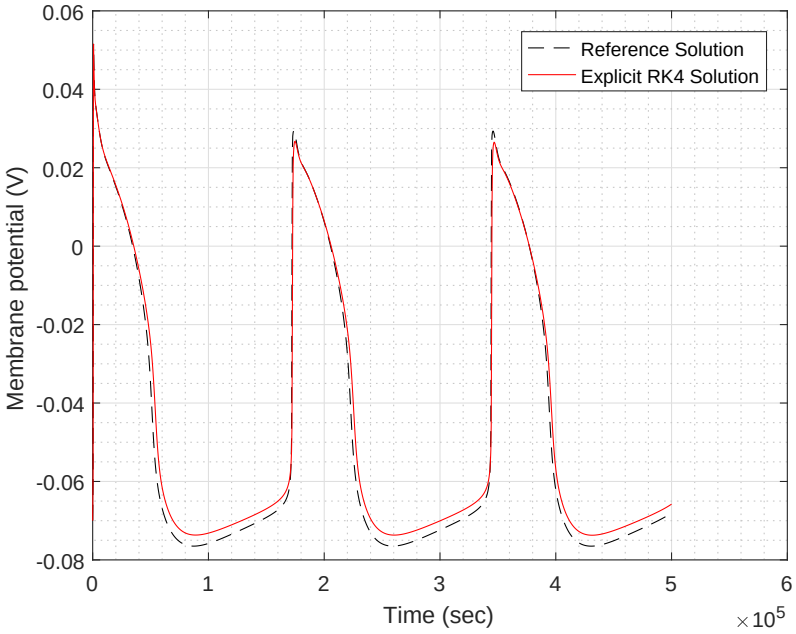


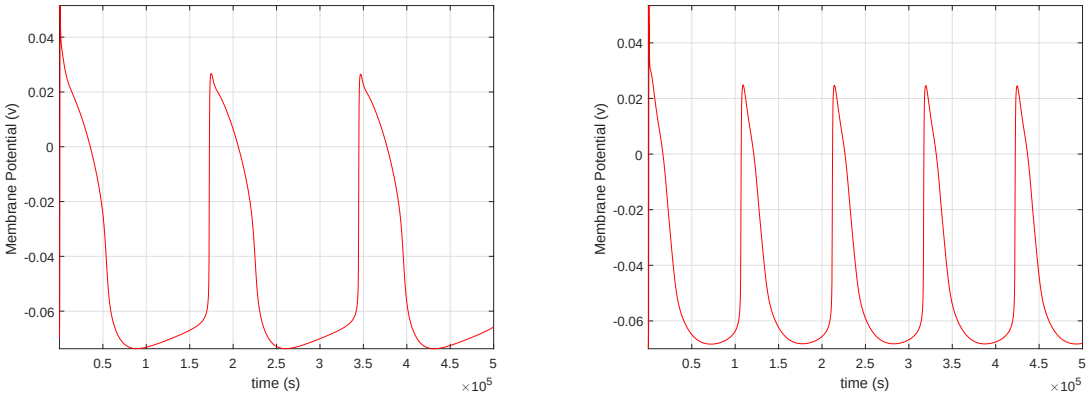
Figure 3: Schematic diagram of the model.

The code has been also implemented in FORTRAN in order to be in accordance with the coding language used in the simulation code for high performance computational mechanics, Alya. Coding in MATLAB has been done in order to verify the correctness of the equations and results obtained from FORTRAN.

Results depicting the behavior of different parameters of interest are provided in the next section.

3 Results and discussion

Figure4 shows the action potential for ventricular-like and atrial-like models. As recent investigations declare, the ventricular-like action potential is more similar to human cardiomyocytes.



(a) Ventricular-like action potential (b) Atrial-like action potential

Figure 4: Membrane Potential

This type of action potential basically consists of 5 main phases, phases 0-4, namely known as rapid upstroke, early repolarization, plateau, late repolarization, and diastole, respectively [14]. The cycle starts by a sharp increase in a very short time in the potential, Phase 0, the potential then reaches its peak at around 0.03 (mV) in the steady state, Phase 1. The third phase, Phase 2, refers to where the potential starts to decrease at a smooth slope. The forth phase, Phase 3, starts at the point where the potential is experiencing sharp decrease compared to the previous phase. The cycle finally ends with an increase in the potential referring to the fifth phase, Phase 4. The phases in a complete cycle for the ventricular-like action potential are depicted in Figure5 for better understanding.

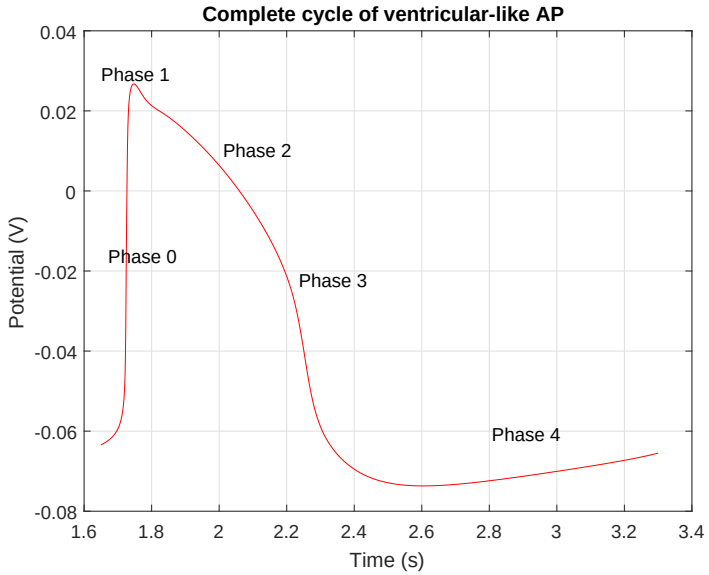
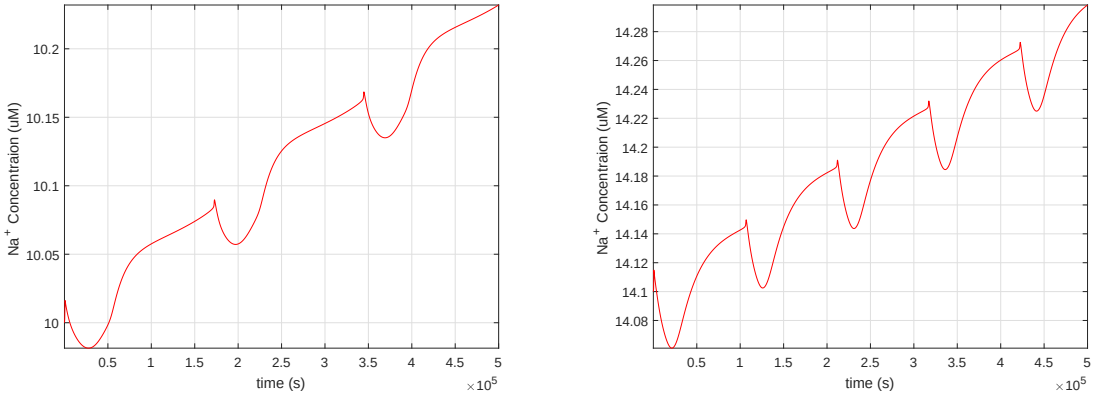


Figure 5: Phases in a cardiac cycle

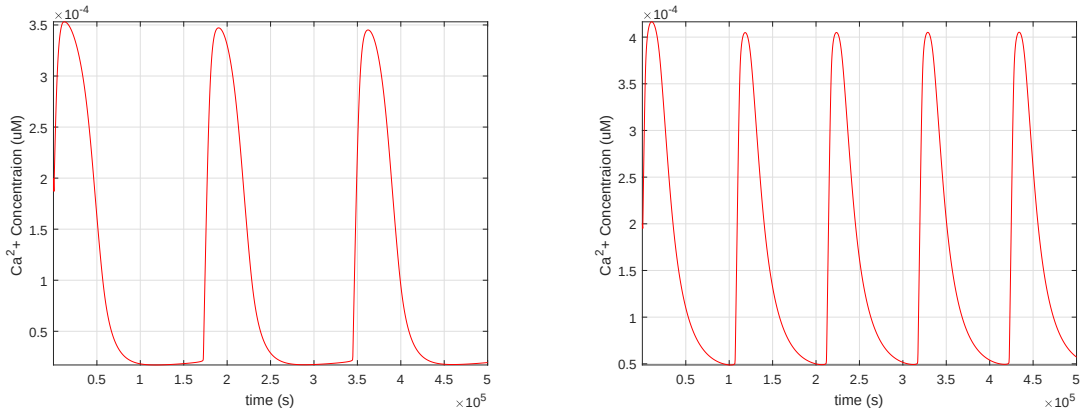
Comparing the two types of action potential, it can be seen that the potential jumps from Phase 1 to Phase 2 in a very short time in atrial-like phenotype, Figure4b, which is potentially the point of difference between atrial-like AP and human CM.

The concentration of two effective ions can be observed in Figure6 and Figure7 for both cell types.



(a) Ventricular-like (b) Atrial-like action

Figure 6: Intracellular Na^+ concentration

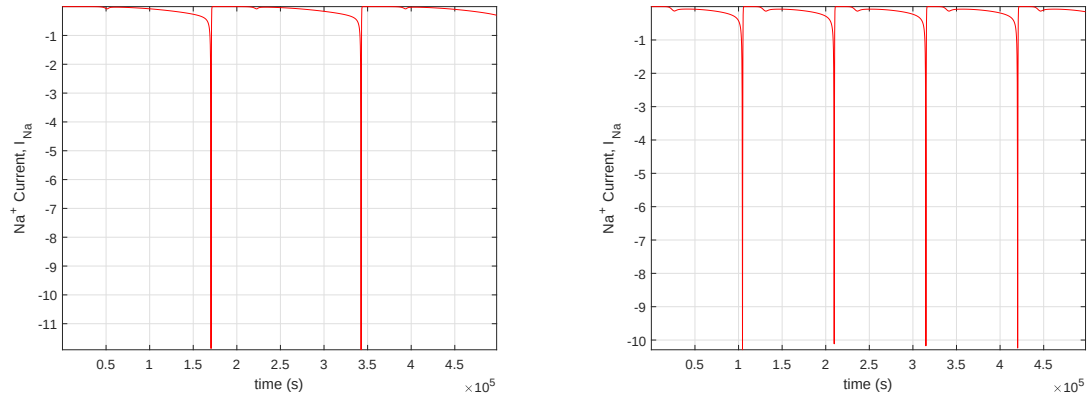


(a) Ventricular-like (b) Atrial-like

Figure 7: Intracellular Ca^{2+} concentration

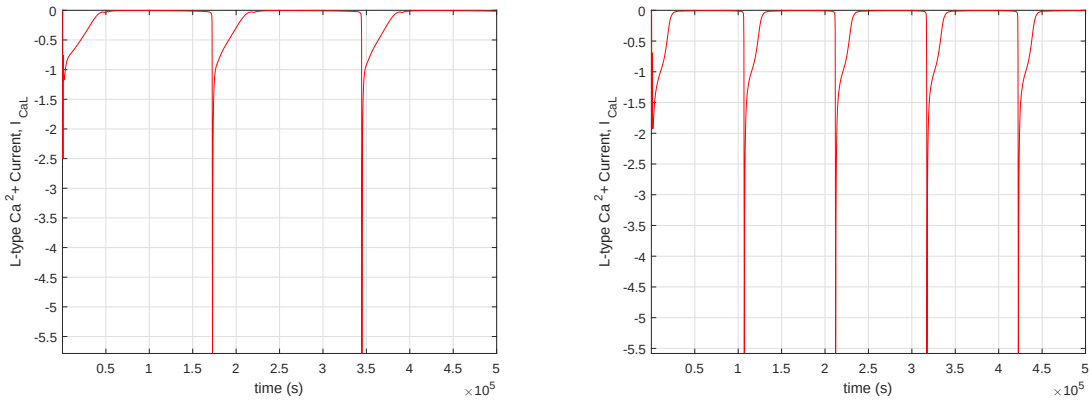
Observing the graphs, it is noticeable that the model as it is implemented requires a long time to achieve steady state. As depicted in Figure6, the results have not reached the steady state; therefore, letting the model run for a lot more than 5 seconds is required. Afterwards, when reaching the steady state, the ODE values can be employed as initial conditions so that there is no need to run the model to steady state every time.

The ion currents which are of great interest in cardiac simulations and investigations are plotted as shown in Figure8 to Figure14.



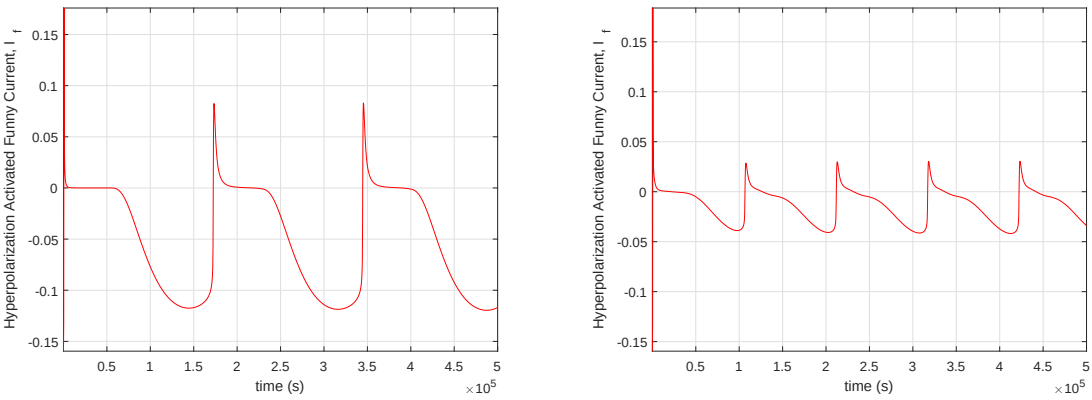
(a) Ventricular-like (b) Atrial-like

Figure 8: Sodium current



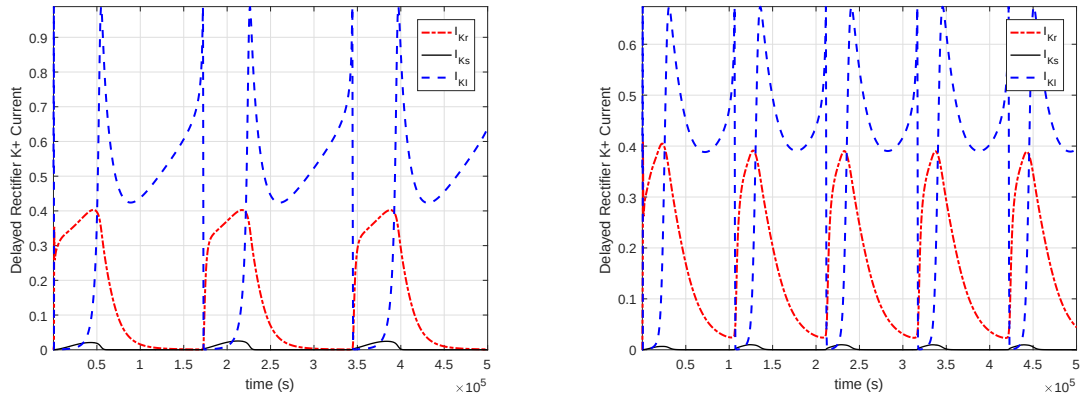
(a) Ventricular-like (b) Atrial-like

Figure 9: L-type Calcium current



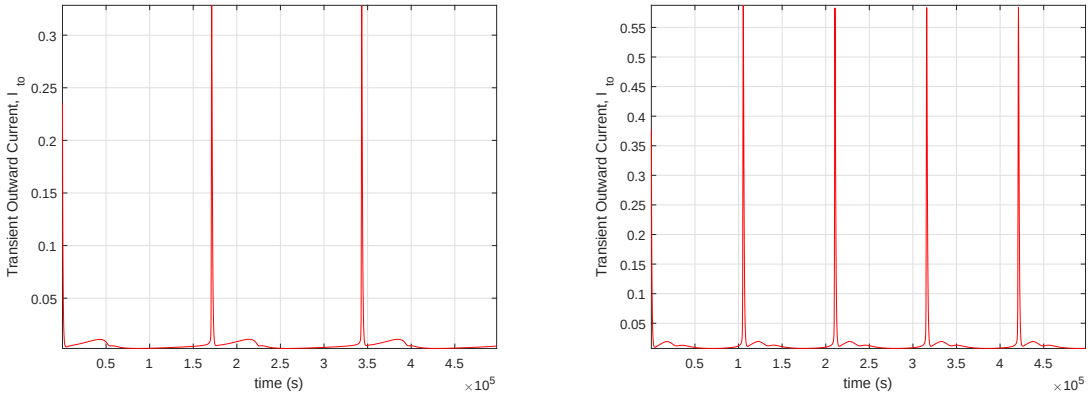
(a) Ventricular-like (b) Atrial-like

Figure 10: Hyperpolarization-activated pacemaker current



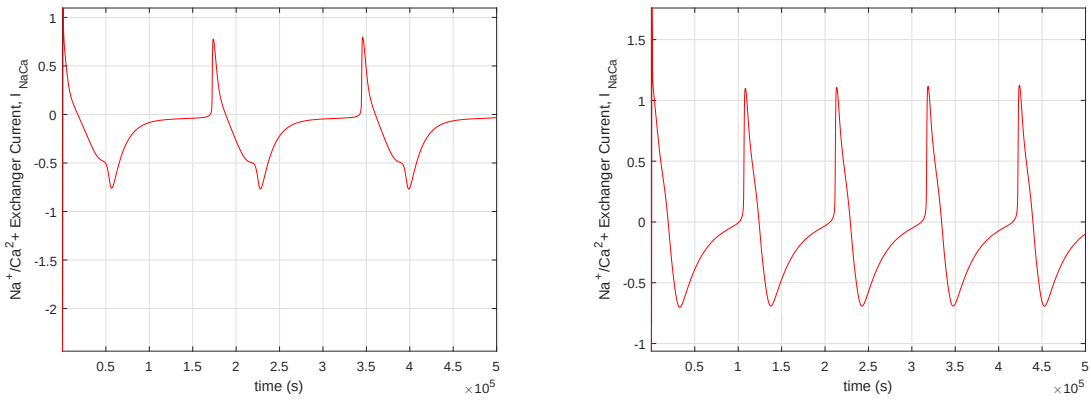
(a) Ventricular-like (b) Atrial-like

Figure 11: Potassium current



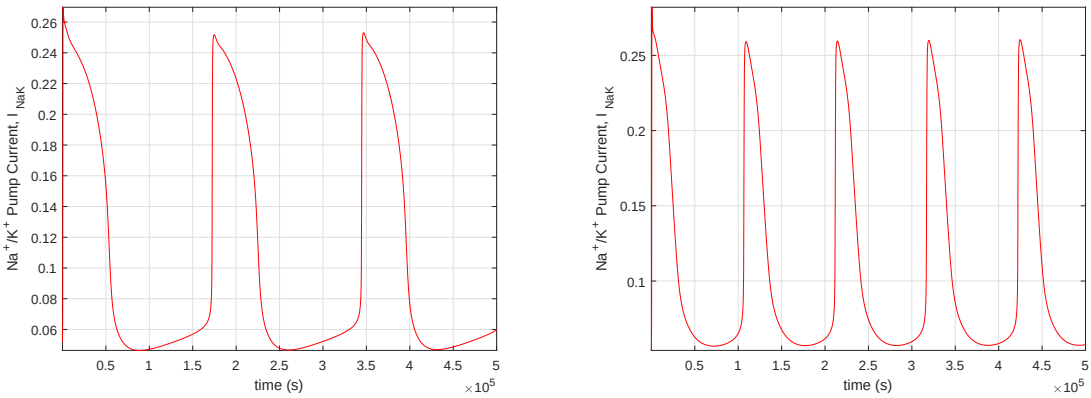
(a) Ventricular-like (b) Atrial-like

Figure 12: Transient outward potassium current



(a) Ventricular-like (b) Atrial-like

Figure 13: Na^+/Ca^{2+} exchanger current



(a) Ventricular-like (b) Atrial-like

Figure 14: Na^+/K^+ pump current

Pluripotent stem cell cardiomyocytes have prominent Na^+ currents with activation and inactivation gating characteristics known as I_{Na}, Figure8.

In ventricular-like hiPSC-CMs, the presence of hyperpolarization-activated I_f promotes phase 4 depolarization and thus contribute to automaticity, Figure10.

Three K^+ currents (I_{to}, I_{Kr}, and I_{Ks}) have been recorded in hiPSC-CMs with maximum densities and activation properties, Figure11. I_{Kr} contributes to repolarization of the cardiac AP, and block of I_{Kr} prolongs the ventricular AP.[14]

As it can be seen in all the ionic currents, the peaks of graphs happen at the same time as the voltage peak. This is due to the fact that all the ionic currents highly depend on the action potential in terms of mathematical equations along with the electrophysiological behavior.

4 Conclusion

Computational modeling is becoming increasingly important in the areas of biology and physiology. The correct implementation of the mathematical models published in the literature is the key point for further application of such models within large scale predictive simulations. In the current work, the goal of implementing an electrophysiological hiPSC-CM model from a printed copy of an 18-ordinary equation model from the paper published by Paci et al. [7] is achieved. The model is tested and analyzed using MATLAB. Afterwards, it is re-written in Fortran language for it to be included within the Alya software.

Taking into account the numerical aspect of the study, the explicit fourth order method of Runge-Kutta is chosen to solve the set of equations. The method is proved to be adequate for application of the model to further predictive simulations. In order to investigate the accuracy of solution suggested by the aforementioned method, two kinds of errors are introduced both of which are of low order from a numerical perspective according to the analysis of the time step. The results depict a small, negligible error for the employed time step of up to $2 \cdot 10^{-5}$ seconds.

Considering the biological aspect of the study, the ventricular-like action potential appears to be more similar to that of human cardiomyocytes based on the data provided in literature. The solution of the mathematical equations exactly simulate the five phases of a complete cardiac cycle. The various ion currents affecting the analysis of hiPSC-CM are taken into consideration since they play a crucial role in the future analysis of simulations related to the diagnostic and treatment of cardiac arrhythmia.

5 Future Perspective

The application of this hiPSC-CM model for bioengineering studies will include the validation of the mathematical model against a wide range of experimental measurements to optimize its parameterization for pre-clinical applications. The subsequent step will be to employ computer simulations to capture the effect of drugs within a large dataset of patient-specific monolayers to assess drug action and efficacy. HiPSC-CM have been employed in an attempt to restore muscular function after myocardial infarction. Finite element models can be employed to study this kind of applications.

6 References

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Appendix

The appendix provides the computational models of ventricular-like and atrial-like human induced pluripotent stem cell derived cardiomyocytes.

Extracellular and intracellular ionic concentrations

$Na_o = 151\ (mM)$
$K_o = 5.4\ (mM)$
$Ca_o = 1.8\ (mM)$
$K_i = 150\ (mM)$

Cell size and dimensions

$$C_m = \begin{cases} 98.7109e^{-12}\ (F), & Ventricular-like \\ 78.6671e^{-12}\ (F), & Atrial-like \end{cases}$$

$$V_c = \begin{cases} 8800\ (um^3), & Ventricular-like \\ 7012\ (um^3), & Atrial-like \end{cases}$$

$$V_{sr} = \begin{cases} 583.73\ (um^3), & Ventricular-like \\ 465.20\ (um^3), & Atrial-like \end{cases}$$

Maximum conductances and currents

$$g_{Na} = \begin{cases} 3.6712302e^3\ (S/F), & Ventricular-like \\ 6.646185e^3\ (S/F), & Atrial-like \end{cases}$$

$g_{CaL} = 8.635702e^{-5}\ (m^3/(F \times s))$

$$g_{to} = \begin{cases} 29.9038\ (S/F), & Ventricular-like \\ 59.8077\ (S/F), & Atrial-like \end{cases}$$

$g_{Kr} = 29.8667\ (S/F)$

$g_{Ks} = 2.041\ (S/F)$

$$g_{K1} = \begin{cases} 28.1492\ (S/F), & Ventricular-like \\ 19.1925\ (S/F), & Atrial-like \end{cases}$$

$g_f = 30.10312\ (S/F)$

$$P_{NaK} = \begin{cases} 1.841424\ (A/F), & Ventricular-like \\ 1.4731392\ (A/F), & Atrial-like \end{cases}$$

$$K_{NaCa} = \begin{cases} 4900\ (A/F), & Ventricular-like \\ 2450\ (A/F), & Atrial-like \end{cases}$$

$a_{rel} = 16.464\ (mM/s)$

$b_{rel} = 0.25\ (mM)$

$c_{rel} = 8.232\ (mM/s)$

$V_{max_{up}} = \begin{cases} 0.56064 : (mM/s), & Ventricular - like \\ 0.22\ (mM/s), & Atrial - like \end{cases}$

$V_{leak} = 4.4444e^{-4}\ (1/s)$

$g_{pCa} = 0.4125\ (A/F)$

$g_{bNa} = 0.9\ (S/F)$

$g_{bCa} = 0.69264\ (S/F)$

Other constants

$Buf_c = 0.25\ (mM)$

$Buf_{sr} = 10\ (mM)$

$K_{Buf_c} = 0.001\ (mM)$

$K_{Buf_{sr}} = 0.3\ (mM)$

$K_{up} = 0.00025\ (mM)$

$K_{pCa} = 0.0005\ (mM)$

$F = 96485.3415\ (C/M)$

$R = 8.314472\ (J/(M \times K))$

$T = 310\ (K)$

$L_0 = 0.025\ (dimensionless)$

$P_{kna} = 0.03\ (dimensionless)$

$K_{sat} = 0.1\ (dimensionless)$

$Km_{Ca} = 1.38\ (mM)$

$Km_{Nai} = 87.5\ (mM)$

$\alpha = 2.8571432\ (dimensionless)$

$\gamma = 0.35\ (dimensionless)$

$K_{mNa} = 40\ (mM)$

$K_{mk} = 1\ (mM)$

Initial conditions

$h_0 = 0.75\ (dimensionless)$

$j_0 = 0.75\ (dimensionless)$

$m_0 = 0\ (dimensionless)$

$d_0 = 0\ (dimensionless)$

$fCa_0 = 1\ (dimensionless)$

$f_{1,0} = 1\ (dimensionless)$

$f_{2,0} = 1\ (dimensionless)$

$r_0 = 0\ (dimensionless)$

$q_0 = 1\ (dimensionless)$

$Xr1_0 = 0\ (dimensionless)$

$Xr2_0 = 1\ (dimensionless)$

$Xs_0 = 0\ (dimensionless)$

$Xf_0 = 0.1\ (dimensionless)$

$g_0 = 1\ (dimensionless)$

$V_0 = -0.07\ (V)$

$Ca_{i_0} = 0.0002\ (mM)$

$Ca_{sr_0} = 0.3\ (mM)$

$$Na_{i_0} = \begin{cases} 10\ (mM), & Ventricular-like \\ 14.1\ (mM), & Atrial-like \end{cases}$$

Membrane Potential

$$\frac{dV}{dtime} = -I_{ion} = -(I_{K1} + I_{to} + I_{Kr} + I_{Ks} + I_{CaL} + I_{NaK} + I_{Na} + I_{NaCa} + I_{pCa} + I_f + I_{bNa} + I_{bCa})$$

Na^+ current, I_{Na}

$$I_{Na} = g_{Na} \cdot m^3 \cdot h \cdot j \cdot (V - E_{Na})$$

I_{Na}, h gate

$$h_{inf} = \frac{1}{\sqrt{1 + e^{\frac{V \times 1000 + 72.1}{5.7}}}}$$

$$\alpha_h = \begin{cases} 0.057 \cdot e^{-\frac{V \times 1000 + 80}{6.8}}, & if\ V < -0.04 \\ 0, & otherwise \end{cases}$$

$$\beta_h = \begin{cases} 2.7 \cdot e^{0.079 \times V \times 1000} + 3.1 \times 10^5 \cdot e^{0.3485 \times V \times 1000}, & if\ V < -0.04 \\ \dfrac{0.77}{0.13 \cdot (1 + e^{\frac{1000 \times V + 10.66}{-11.1}})}, & otherwise \end{cases}$$

$$\tau_h = \begin{cases} \dfrac{1.5}{(\alpha_h + \beta_h) \times 1000}, & if\ V < -0.04 \\ 2.542e^{-3}, & otherwise \end{cases}$$

$$\frac{dh}{dtime} = \frac{h_{inf} - h}{\tau_h}$$

$\mathbf{I_{Na}}$, j gate

$$j_{inf} = h_{inf}$$

$$\alpha_j = \begin{cases} \dfrac{(V \times 1000 + 37.78) \cdot (-25428 \cdot e^{0.2444 \times V \times 1000} - 6.948 \cdot 10^{-6} \cdot e^{-0.04391 \times V \times 1000})}{1 + e^{0.311 \cdot (V \times 1000 + 79.23)}}, & if\ V < -0.04 \\ 0, & otherwise \end{cases}$$

$$\beta_j = \begin{cases} \dfrac{0.02424 \cdot e^{-0.01052 \times V \times 1000}}{1 + e^{-0.1378 \cdot (V \times 1000 + 40.14)}}, & if\ V < -0.04 \\ \dfrac{0.6 \cdot e^{0.057 \cdot V \times 1000}}{1 + e^{-0.1 \cdot (V \times 1000 + 32)}}, & otherwise \end{cases}$$

$$\tau_j = \frac{7}{(\alpha_j + \beta_j) \times 1000}$$

$$\frac{dj}{dtime} = \frac{j_{inf} - j}{\tau_j}$$

$\mathbf{I_{Na}}$, m gate

$$m_{inf} = \frac{1}{\left(1 + e^{\frac{-34.1 - V \times 1000}{5.9}}\right)^{\frac{1}{3}}}$$

$$\alpha_m = \frac{1}{1 + e^{\frac{-60 - V \times 1000}{5}}}$$

$$\beta_m = \frac{0.1}{1 + e^{\frac{V \times 1000 + 35}{5}}} + \frac{0.1}{1 + e^{\frac{V \times 1000 - 50}{200}}}$$

$$\tau_m = \frac{\alpha_m \cdot \beta_m}{1000}$$

$$\frac{dm}{dtime} = \frac{m_{inf} - m}{\tau_m}$$

L–Type $\mathbf{Ca^{2+}}$ current, $\mathbf{I_{CaL}}$

$$I_{CaL} = \frac{g_{CaL} \cdot 4 \cdot V \cdot F^2 \cdot (Ca_i \cdot e^{\frac{2 \cdot V \cdot F}{R \cdot T}} - 0.341 \cdot Ca_o) \cdot d \cdot f_1 \cdot f_2 \cdot f_{Ca}}{R \cdot T \cdot (e^{\frac{2 \cdot V \cdot F}{R \cdot T}} - 1)}$$

$\mathbf{I_{CaL}}$, d gate

$$d_{inf} = \begin{cases} \dfrac{1}{1+e^{-\frac{V\times 1000+9.1}{7}}}, & Ventricular-like \\ \dfrac{1}{1+e^{-\frac{V\times 1000+5.986}{7}}}, & Atrial-like \end{cases}$$

$$\alpha_d = 0.25 + \frac{1.4}{1+e^{\frac{-35-V\times 1000}{13}}}$$

$$\beta_d = \frac{1.4}{1+e^{\frac{5+V\times 1000}{5}}}$$

$$\gamma_d = \frac{1}{1+e^{\frac{50-V\times 1000}{20}}}$$

$$\tau_d = \frac{\alpha_d \cdot \beta_d + \gamma_d}{1000}$$

$$\frac{dd}{dtime} = \frac{d_{inf} - d}{\tau_d}$$

$\mathbf{I_{CaL}}$, $\mathbf{f_{Ca}}$ gate

$$\alpha_{f_{Ca}} = \frac{1}{1+\left(\frac{Ca_i}{0.0006}\right)^8}$$

$$\beta_{f_{Ca}} = \frac{0.1}{1+e^{\frac{Ca_i-0.0009}{0.0001}}}$$

$$\gamma_{f_{Ca}} = \frac{0.3}{1+e^{\frac{Ca_i-0.00075}{0.0008}}}$$

$$f_{Ca_{inf}} = \frac{\alpha_{f_{Ca}} + \beta_{f_{Ca}} + \gamma_{f_{Ca}}}{1.3156}$$

$$\tau_{f_{Ca}} = 0.002\ (second)$$

$$const_{f_{Ca}} = \begin{cases} 0, & if\ f_{Ca_{inf}} > f_{Ca}\ and\ V > -0.06 \\ 1, & otherwise \end{cases}$$

$$\frac{df_{Ca}}{dtime} = const_{f_{Ca}} \cdot \frac{f_{Ca_{inf}} - f_{Ca}}{\tau_{f_{Ca}}}$$

$\mathbf{I_{CaL}}$, $\mathbf{f_1}$ gate

$$f_{1inf} = \begin{cases} \dfrac{1}{1+e^{\frac{V\times 1000+26}{3}}}, & Ventricular-like \\ \dfrac{1}{1+e^{\frac{V\times 1000+25.226}{3}}}, & Atrial-like \end{cases}$$

$$\tau_{f1} = \left(1102.58 \cdot e^{-\left[\frac{(V\times 1000+27)^2}{15}\right]^2} + \frac{200}{1+e^{\frac{13-V\times 1000}{10}}} + \frac{180}{1+e^{\frac{30+V\times 1000}{10}}} + 20\right) \begin{cases} \frac{1+1433(Ca_i-50e^{-6})}{1000}, & if\ \frac{df_1}{dtime} > 0 \\ 0.001, & otherwise \end{cases}$$

$$\frac{df_1}{dtime} = \frac{f_{1inf} - f_1}{\tau_{f1}}$$

I_{CaL}, f_2 gate

$$f_{2inf} = \begin{cases} \dfrac{0.67}{1 + e^{\frac{V \times 1000 + 35}{4}}} + 0.33, & Ventricular - like \\ \dfrac{0.67}{1 + e^{\frac{V \times 1000 + 31.226}{4}}} + 0.33, & Atrial - like \end{cases}$$

$$\tau_{f1} =$$

$$\left(600 \cdot (e^{-\frac{(V \times 1000 + 25)^2}{170}}) + \frac{31}{1 + e^{\frac{25 - V \times 1000}{10}}} + \frac{16}{1 + e^{\frac{30 + V \times 1000}{10}}}\right) \begin{cases} 0.001, & Ventricular - like \\ 0.002, & Atrial - like \end{cases}$$

$$\frac{df_2}{dtime} = \frac{f_{2inf} - f_2}{\tau_{f2}}$$

Transient outward current, I_{to}

$$I_{to} = g_{to} \cdot r \cdot q \cdot (V - E_K)$$

I_{to}, r gate

$$r_{inf} = \frac{1}{1 + e^{\frac{22.3 - V \times 1000}{18.75}}}$$

$$\tau_r = \left(\frac{14.405516}{1.037\ e^{0.09\ (V \times 1000 + 30.61)} + 0.369\ e^{-0.12\ (V \times 1000 + 23.84)}} + 2.75352\right) \times 10^{-3}$$

$$\frac{dr}{dtime} = \frac{r_{inf} - r}{\tau_r}$$

I_{to}, q gate

$$q_{inf} = \frac{1}{1 + e^{\frac{53 + V \times 1000}{13}}}$$

$$\tau_q = \left(\frac{39.102}{0.57\ e^{-0.08\ (V \times 1000 + 44)} + 0.065\ e^{0.1\ (V \times 1000 + 45.93)}} + 6.06\right) \times 10^{-3}$$

$$\frac{dq}{dtime} = \frac{q_{inf} - q}{\tau_q}$$

Rapid delayed rectifier K^+ current, I_{Kr}

$$I_{Kr} = g_{Kr} \cdot \sqrt{\frac{K_o}{5.4}} \cdot Xr_1 \cdot Xr_2 \cdot (V - E_K)$$

I_{Kr}, Xr_1 gate

$$V_{1/2} = 1000 \cdot \left(-\frac{RT}{FQ} \cdot ln\left(\frac{(1 + \frac{Ca_o}{2.6})^4}{L_0 \cdot (1 + \frac{Ca_o}{0.58})^4}\right) - 0.019\right)$$

$$xr1_{inf} = \frac{1}{1 + e^{\frac{V_{1/2} - V \times 1000}{4.9}}}$$

$$\alpha_{xr1} = \frac{450}{1 + e^{\frac{-45 - V \times 1000}{10}}}$$

$$\beta_{xr1} = \frac{6}{1 + e^{\frac{30 + V \times 1000}{11.5}}}$$

$$\tau_{xr1} = \alpha_{xr1} \cdot \beta_{xr1} \cdot 10^{-3}$$

$$\frac{dXr_1}{dtime} = \frac{xr1_{inf} - Xr_1}{\tau_{xr1}}$$

$\mathbf{I_{Kr}}$, $\mathbf{Xr_2}$ gate

$$xr2_{inf} = \frac{1}{1 + e^{\frac{V \times 1000 + 88}{50}}}$$

$$\alpha_{xr2} = \frac{3}{1 + e^{\frac{-V \times 1000 - 60}{20}}}$$

$$\beta_{xr2} = \frac{1.12}{1 + e^{\frac{V \times 1000 - 60}{20}}}$$

$$\tau_{xr2} = \alpha_{xr2} \cdot \beta_{xr2} \cdot 10^{-3}$$

$$\frac{dXr_2}{dtime} = \frac{xr2_{inf} - Xr_2}{\tau_{xr2}}$$

Slow delayed rectifier $\mathbf{K^+}$ current, $\mathbf{I_{Ks}}$

$$I_{Ks} = g_{Ks} \cdot Xs^2 \cdot \left(1 + \frac{0.6}{1 + \left(\frac{3.8 \cdot 10^{-5}}{Ca_i}\right)^{1.4}}\right) \cdot (V - E_{Ks})$$

$\mathbf{I_{Ks}}$, Xs gate

$$Xs_{inf} = \frac{1}{1 + e^{\frac{-V \times 1000 - 20}{16}}}$$

$$\alpha_{xs} = \frac{1100}{\sqrt{1 + e^{\frac{-V \times 1000 - 10}{6}}}}$$

$$\beta_{xs} = \frac{1}{1 + e^{\frac{V \times 1000 - 60}{20}}}$$

$$\tau_{xs} = \alpha_{xs} \cdot \beta_{xs} \cdot 10^{-3}$$

$$\frac{dXs}{dtime} = \frac{Xs_{inf} - Xs}{\tau_{xs}}$$

Inward rectifier $\mathbf{K^+}$ current, $\mathbf{I_{K1}}$

$$\alpha_{K_1} = \frac{3.91}{1 + e^{0.5942\ (1000\times(V-E_K)-200)}}$$

$$\beta_{K_1} = \frac{-1.509\ e^{0.0002\ (1000\times(V-E_K)+100)} + e^{0.5886\ (1000\times(V-E_K)-10)})}{1 + e^{0.4547\times1000\times(V-E_K)}}$$

$$xK1_{inf} = \frac{\alpha_{K_1}}{\alpha_{K_1} + \beta_{K_1}}$$

$$I_{K1} = g_{K1} \cdot xK1_{inf} \cdot \sqrt{\frac{K_o}{5.4}} \cdot (V - E_K)$$

Hyperpolarization activated funny current, $\mathbf{I_f}$

$$I_f = g_f \cdot Xf \cdot (V - E_f)$$

$\mathbf{I_f}$, $\mathbf{X_f}$ gate

$$xf_{inf} = \frac{1}{1 + e^{\frac{V\times1000+77.85}{5}}}$$

$$\tau_{xf} = \frac{1900\ e^{-3}}{1 + e^{\frac{V\times1000+15}{10}}}$$

$$\frac{dXf}{dtime} = \frac{xf_{inf} - Xf}{\tau_{xf}}$$

Na^+ / K^+ pump current, $\mathbf{I_{NaK}}$

$$I_{NaK} = \frac{\frac{P_{NaK}\cdot K_o \cdot Na_i}{K_o + K_{mk}}}{Na_i + K_{mNa}} \over 1 + 0.1245\ e^{-0.1\cdot\frac{VF}{RT}} + 0.353\ e^{-\frac{VF}{RT}}$$

Na^+ / Ca^{2+} exchanger current, $\mathbf{I_{NaCa}}$

$$I_{NaCa} = \frac{K_{NaCa} \cdot \left(e^{\gamma\cdot\frac{V\cdot F}{R\cdot T}} \cdot Na_i^3 \cdot Ca_o - e^{(\gamma-1)\cdot\frac{V\cdot F}{R\cdot T}} \cdot Na_i^3 \cdot Ca_o \cdot \alpha\right)}{((Km_{Na_i})^3 + Na_o^3) \cdot (Km_{Ca} + Ca_o)\left(1 + K_{sat} \cdot e^{(\gamma-1)\cdot\frac{V\cdot F}{R\cdot T}}\right)}$$

Ca^{2+} dynamics

$$I_{rel} = \left(\frac{a_{rel} \cdot {Ca_{SR}}^2}{{b_{rel}}^2 + {Ca_{SR}}^2} + c_{rel}\right) \cdot d \cdot g \begin{cases} 0.0411, & Ventricular - like \\ 0.0556, & Atrial - like \end{cases}$$

$$I_{up} = \frac{V_{max_{up}}}{1 + \frac{{K_{up}}^2}{{Ca_i}^2}}$$

$$I_{leak} = V_{leak} \cdot (Ca_{SR} - Ca_i)$$

$$g_{inf} = \begin{cases} \dfrac{1}{1 + \left(\frac{Ca_i}{0.00035}\right)^6}, & if\ Ca_i \leq 0.00035 \\ \dfrac{1}{1 + \left(\frac{Ca_i}{0.00035}\right)^{16}}, & otherwise \end{cases}$$

$$constii = \begin{cases} 0, & if\ g_{inf} > g\ and\ V > -0.06 \\ 1, & otherwise \end{cases}$$

$$\tau_g = 0.002$$

$$\frac{dg}{dtime} = constii \cdot \frac{g_{inf} - g}{\tau_g}$$

$$\frac{dg}{dtime} = constii \cdot \frac{g_{inf} - g}{\tau_g}$$

$$Ca_{i_{bufc}} = \frac{1}{1 + \frac{buf_c \cdot K_{buf_c}}{\left(Ca_i + K_{buf_c}\right)^2}}$$

$$Ca_{sr_{bufsr}} = \frac{1}{1 + \frac{buf_{sr} \cdot K_{buf_{sr}}}{\left(Ca_{SR} + K_{buf_{sr}}\right)^2}}$$

$$\frac{dCa_i}{dtime} = Ca_{i_{bufc}} \cdot \left(I_{leak} - I_{up} + I_{rel} - \frac{I_{CaL} + I_{bCa} + I_{pCa} - 2 \cdot I_{NaCa}}{2 \cdot V_c \cdot F \cdot e^{-18}} \cdot C_m\right)$$

$$\frac{dCa_{SR}}{dtime} = \frac{Ca_{sr_{bufsr}} \cdot V_c}{V_{sr}} \cdot (I_{up} - (I_{rel} + I_{leak}))$$

Ca^{2+} pump current, I_{pCa}

$$I_{pCa} = \frac{g_{pCa} \cdot Ca_i}{Ca_i + K_{pCa}}$$

Reversal potentials

$$E_{Na} = \frac{R \cdot T}{F} \cdot \ln \frac{Na_o}{Na_i}$$

$$E_K = \frac{R \cdot T}{F} \cdot \ln \frac{K_o}{K_i}$$

$$E_{Ks} = \frac{R \cdot T}{F} \cdot \ln \frac{K_o + P_{kna} \cdot Na_o}{K_i + P_{kna} \cdot Na_i}$$

$$E_{Ca} = \frac{0.5 \cdot R \cdot T}{F} \cdot \ln \frac{Ca_o}{Ca_i}$$

$$E_f = -0.017$$

Sodium dynamics

$$\frac{dNa_i}{dtime} = -C_m \cdot \frac{I_{Na} + I_{bNa} + 3 \times I_{NaK} + 3 \times I_{NaCa}}{F \cdot V_c \cdot e^{-18}}$$

Background currents

$I_{bNa} = g_{bNa} \cdot (V - E_{Na})$

$I_{bCa} = g_{bCa} \cdot (V - E_{Ca})$