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QUANTITATIVE MODELLING OF EFFECT OF TRANSVERSE-AXIAL TUBULAR SYSTEM ON ELECTRICAL ACTIVITY OF CARDIAC CELLS: DEVELOPMENT OF MODEL

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Abstract: ***The transverse-axial tubular system (TAT-system) of cardiac muscle is a structure that allows rapid propagation of excitation into the cell interior. As suggested in many recent experimental works, it could have a significant effect on cardiac cell function induced by the accumulation or the depletion of ions in restricted tubular space. In our previous work [27], the basic properties of TAT-system were formulated and preliminary simulations characterizing its effect on cellular electrical activity realised. In this article, we describe the design of a more complex model of ventricular myocyte based mostly on data from guinea pig. The model integrates the description of electrical activity of surface and tubular membranes with the detailed description of mechanisms controlling the intracellular and tubular ion concentrations.***

Key words*: cardiac cell, transverse-axial tubular system, quantitative modelling*

1. INTRODUCTION

Mathematical models of excitable cells have undergone a remarkable evolution since Hodgkin and Huxley's pioneering work in the 1950s [1]. The modelling paradigm established by Hodgkin and Huxley uses the total current flowing through ion channels in the membrane to determine the transmembrane potential (V_m) [2, 3, 4, 5]. A later stage in the evolution of mathematical models has been the development of cell models that account for dynamic changes in intracellular ion concentrations [6, 7, 8, 9]. These dynamic models build upon the original paradigm describe not only electrical properties of cellular membrane but also the function of important cytosolic compartments (fuzzy space, sarcoplasmic reticulum) and ion buffers.

Although the current structure of dynamic models has been widely accepted for studying the electromechanical activity of cardiac cells, the new experimental findings strongly suggest the necessity of its further development. One of the phenomena that cannot be simulated and quantified by the mathematical models of current level is the effect of transverse-axial tubular system (TAT-system) on electrical activity of ventricular cardiomyocytes. This phenomenon induced by accumulation and depletion of ions in restricted tubular spaces is considered to be of great significance for cellular arrhythmogenesis [10, 11, 12]. To study this process, we have designed a complex model of ventricular myocyte including description of the TAT-system function. The development of this model is described in the present paper.

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2. METHODS

Generally, the model is based on a quantitative description of electrical activity of ventricular myocyte proposed by Luo and Rudy in 1994 [8]. It includes formulation of the ion transporting systems in surface and tubular membrane as well as description of the sarcoplasmic reticulum (uptake and release compartment) and Ca-buffers. The model also incorporates a subsarcolemal compartment - fuzzy space that proved to be necessary for mechanisms of Ca-release from sarcoplasmic reticulum. A schematic diagram of the model is shown in Fig. 1.

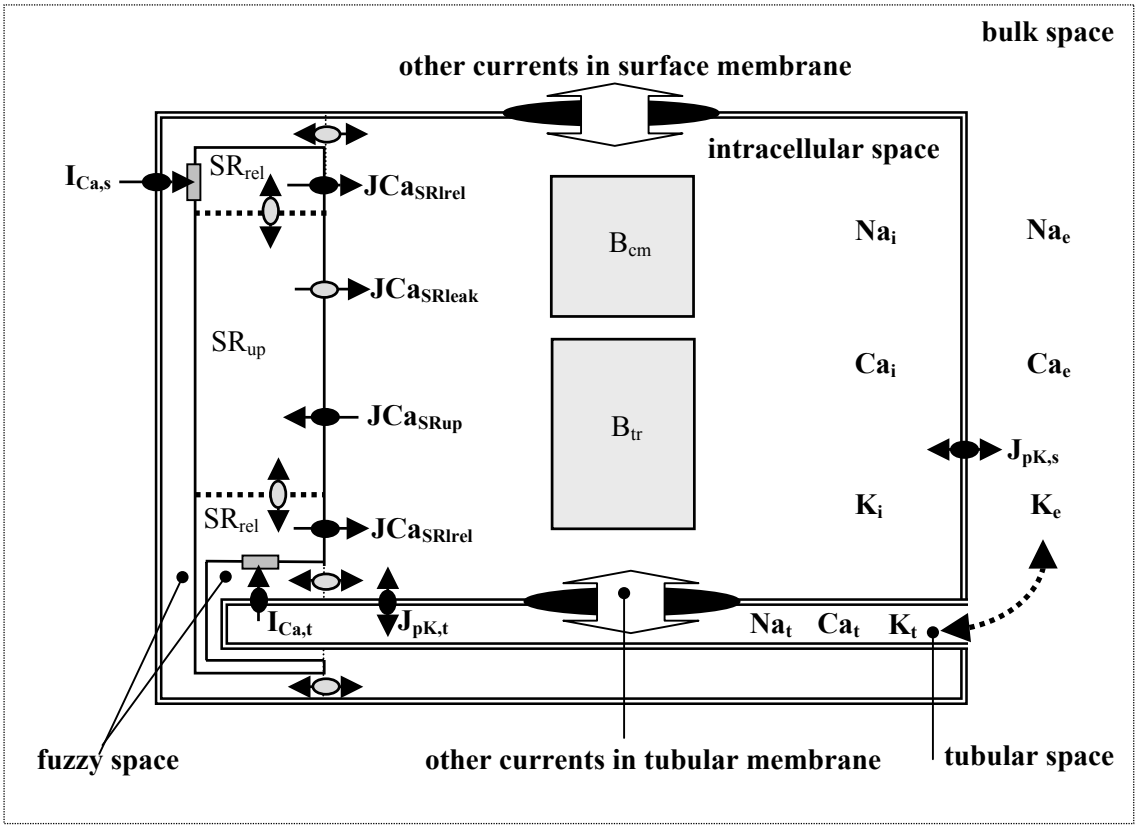

Figure 1. *Schematic diagram of the ventricular cell model. The description of electrical activity of surface and tubular membrane comprises ion currents (I_{Na}, I_{Ca}, I_K, I_{K1}, I_{Kp}, $I_{ns(Ca)}$, $I_{Na,b}$, $I_{Ca,b}$, I_{NaCa}, I_{NaK}, I_{pCa}) as introduced in [8] and electroneutral K-pump flux (J_{pK}) for maintenance of potassium homeostasis. The intracellular space contains Ca-uptake (SR_{up}) and Ca-release (SR_{rel}) compartment of sarcoplasmic reticulum and Ca-buffering by calmodulin (B_{cm}) and troponin (B_{tr}). The small grey rectangles in SR_{rel} membrane denote the ryanodine receptors. The arrows with grey ellipse denote the Ca-diffusion in cytosol and the dashed arrow between tubular and bulk space represents Na, K and Ca-diffusion between these two spaces.*

The model was designed and adjusted for the best fit with experimental data from guinea pig cardiomyocytes. The main modifications of quantitative description with respect to the model proposed by Luo and Rudy (1994) [8] are summarised in the following items:

- The description of I_{Na}-channel gating is restricted to three-activation (m^3) and one inactivation (h) gates. The rate constants for activation (α_m, β_m) and inactivation (α_h, β_h) are formulated according to experimental data in [13] and their resulting form is:

$\alpha_m = 117{,}26 \cdot (V_m + 59{,}25)/(1-\exp(-0{,}55 \cdot (V_m + 59{,}25)))$, $\beta_m = 3800 \cdot \exp(-0{,}0723 \cdot (V_m + 61))$

$\alpha_h = 15{,}518/(1+\exp(0{,}188 \cdot (V_m + 68{,}2)))$, $\beta_h = 18{,}77 \cdot (V_m + 74{,}4)/(1-\exp(-0{,}4 \cdot (V_m + 74{,}4)))$.

■ The description of reversal voltage for I_{Na} includes *12 %* permeability of I_{Na} for potassium ions (according to [7]). Without this term the reversal voltage was too high.

■ The half-saturation concentration for Ca-induced inactivation of I_{Ca} was diminished to *0,5 μM*. The process of inactivation is described as time dependent (with time constant *0,07 s*) to produce correct values of peak intracellular transient at different stimulation frequencies as observed in guinea pig ventricular cardiomyocytes [29].

■ The voltage dependence of I_{Kp} (k_{Kp}) was slightly modified to ensure the characteristic configuration of ventricular action potential. Its modified form is: $k_{Kp}=1/(1+exp(-V_m/6))$.

■ The saturation factor of I_{NaCa} at very negative potentials (k_{sat}) was adjusted to 0,25 to meet the experimental results of [14].

■ The electroneutral potassium pump (J_{pK}) was included into the model to maintain stable intracellular potassium concentration (K_i) for given K_e. The pump was described as K_e and K_i dependent and its form is: $J_{pK} = J_{pKmax} \cdot (K_i/(K_i+140) - K_e/(K_e+5,4))$.

■ The stimulus current was incorporated into the equation controlling the intracellular potassium concentration to comply with the charge conservation principle [15].

■ The subsarcolemal fuzzy space was included into the model to enable close interaction between I_{Ca} and ryanodine receptors. Its volume (*2 %* of cytoplasm [9]) takes into account the function of Ca-buffers in this space [16]. The time constant of diffusion between fuzzy space and cytosol is set according to data in [16] to *0,1 ms*.

■ The Ca-induced Ca release from SR_{rel} (CICR) is completely reformulated on the basis of the description in [9]. The rate constants of this process were modified to produce acceptable Ca_i-transients at different stimulation frequencies. The kinetic scheme of CICR together with its quantitative description is illustrated in Fig. 2.

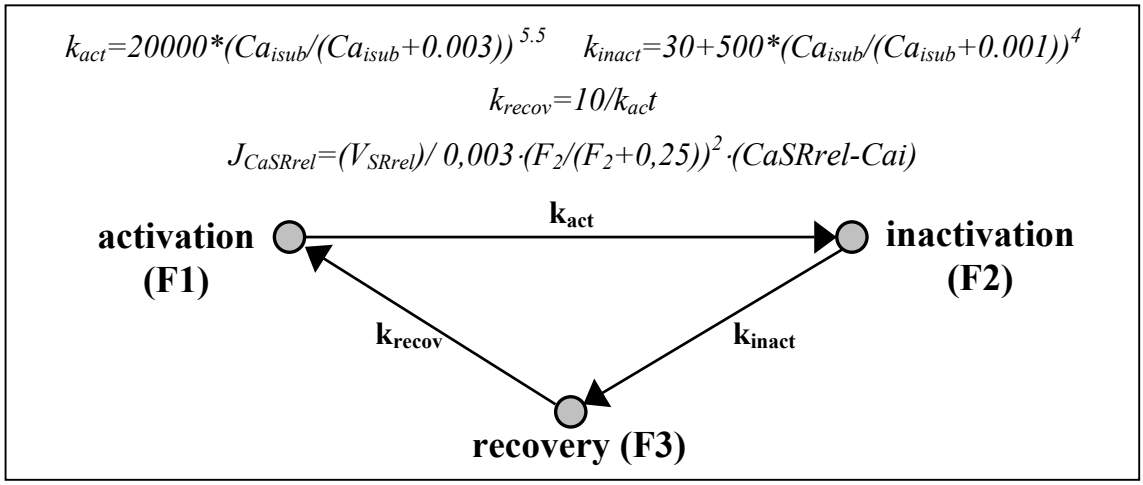

Figure 2. *Kinetic scheme and quantitative description of Ca induced Ca release from sarcoplasmic reticulum release compartment (SR_{rel}).*

The dimensions of the ventricular guinea pig myocytes were assumed to be: *100 μm* length and *10 μm* radius (cylindrical geometry). The proportions of intracellular compartments are in identical ratio as proposed in [8].

The geometric parameters of TAT-system were set to meet the experimental results obtained from rat ventricular myocytes [17]. These parameters comprise density of tubules in surface membrane (*27,78* 10^6 *tubules/cm*^{2}), average radius of tubules ($r_t=127 \cdot 10^{-7}$ *cm)* and their length ($l_t=10\mu m$). The tubular membrane forms *69 %* of total membrane area and the fractional volume of TAT-system is *3,12 %* of cellular volume.

The quantitative descriptions of electrical activity of surface and tubular membrane were interconnected by total series resistance of TAT-system (R_{st}) as shown in Fig. 3.

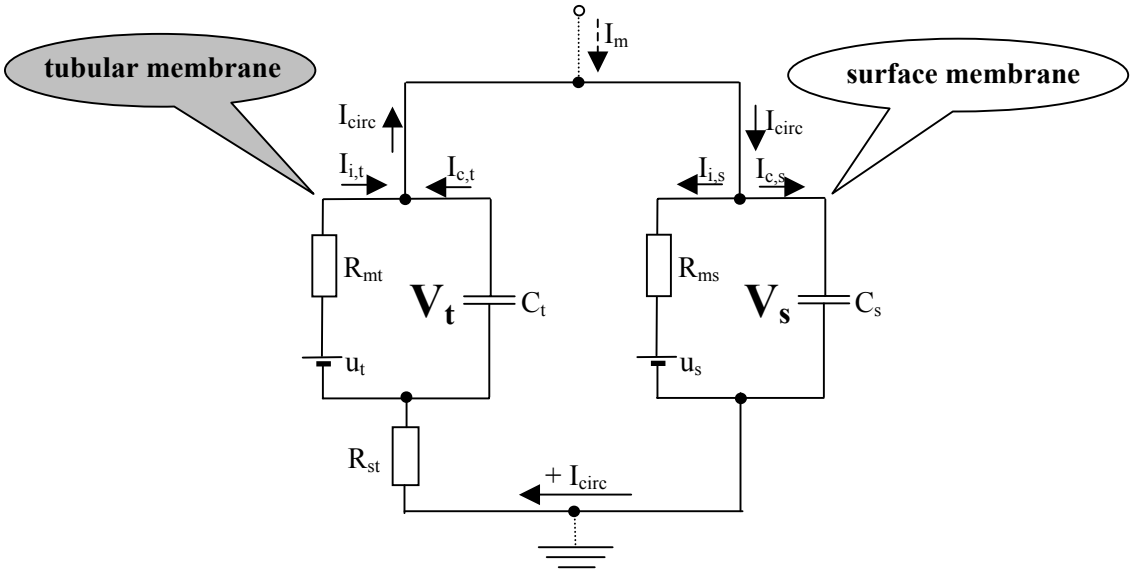

Figure 3. *Scheme showing the electrical interaction between surface and tubular membrane.*

The total series resistance of the TAT-system (R_{st}) was calculated from the equation:

$$R_{st} = 0{,}5 \cdot l_t \cdot R_{ext} / (\pi \cdot r_t^2 \cdot n_t)$$

taking into account the specific resistivity of extracellular solution R_{ext} (for the Tyrode solution $R_{ext} \approx 83{,}33\ \Omega \cdot cm$), average radius of tubules (r_t), effective length of tubules (l_t) and eventually the number of tubules (n_t) for the given area of surface membrane.

The maximum ion conductivities, permeabilities, currents or fluxes of individual transporters in surface and tubular membrane are summarised in table 1.

	unite	surface membrane	tubular membrane	according to
$\mathbf{g_{Na}}$	[mS/cm^2]	30	30	[18, 24]
$\mathbf{g_{K}}$	[mS/cm^2]	0,387	0,0435	[20]
$\mathbf{g_{K1}}$	[mS/cm^2]	0,4838	0,8696	[12]
$\mathbf{g_{Kp}}$	[mS/cm^2]	0,0183	0,0183	
$\mathbf{g_{Na,b}}$	[mS/cm^2]	0,0035	0,0035	
$\mathbf{g_{Ca,b}}$	[mS/cm^2]	0,0015	0,0015	
$\mathbf{P_{Ca}}$	[cm/s]	3,51e-4	3,51e-4	[19, 24]
$\mathbf{P_{ns(Ca)}}$	[cm/s]	1,75e-7	1,75e-7	
$\mathbf{I_{NaCa,max}}$	[µS/cm^2]	1000	1797	[22, 23]
$\mathbf{I_{NaK,max}}$	[µS/cm^2]	1,949	1,949	[25]
$\mathbf{I_{pCa,max}}$	[µS/cm^2]	2,9673	0,3334	[26]
$\mathbf{J_{pK,max}}$	[pM/s/cm^2]	310	310	

Table 1. *Electrical properties of surface and tubular membrane transport systems used in the model.*

The time constants of ion diffusion between TAT-system and bulk solution were slightly modified (with respect to our previous study [27]) to meet the latest experimental data [28, 19]. They are *64 ms* for potassium and sodium ions and *250 ms* for calcium ions.

The model was implemented in the program system MATLAB 5.3 and the numerical computation of 26 differential equations was performed using the solver for stiff systems ODE-15s.

3. RESULTS OF QUANTITATIVE MODELLING

The simulations presented in this section show basic behaviour of the model in response to three stimulation pulses (*8.9 nA*, *1 ms, 1 Hz*) applied at resting state.

Fig. 4 depicts action potentials and main membrane currents in surface and tubular membrane. The different magnitudes of tubular membrane currents (denoted by green line) result from different area of tubular membrane versus surface membrane as well as from different densities of some channels, exchangers or pumps (see Table 1) in tubular membrane.

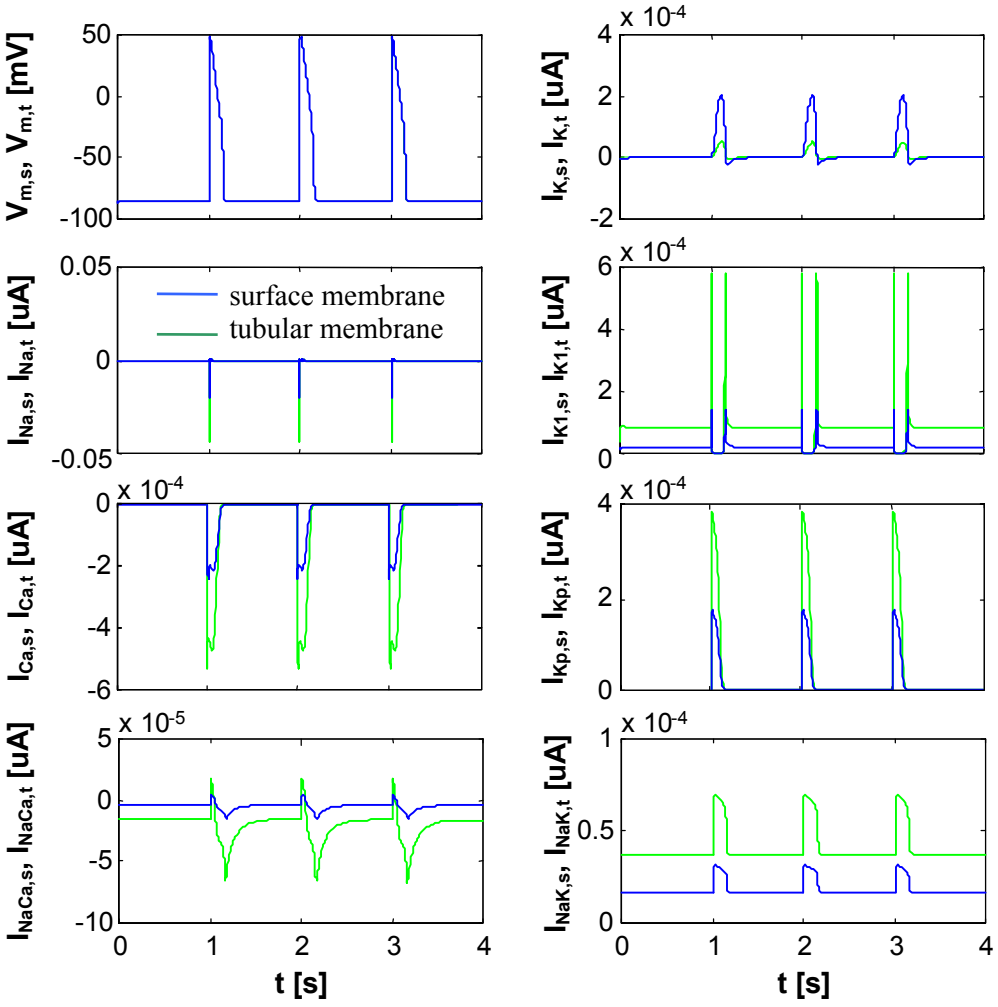

Figure 4. *Action potentials ($V_{m,s}$, $V_{m,t}$) and main ionic currents in surface and tubular membrane in response to three stimulation pulses (8.9 nA, 1 ms, 1 Hz) applied in resting state. The currents here visualised are: fast sodium currents ($I_{Na,s}$, $I_{Na,t}$); calcium current through L-type channels ($I_{Ca,s}$, $I_{Ca,t}$); delayed rectifier potassium currents ($I_{K,s}$, $I_{K,t}$); inwardly rectifying potassium currents ($I_{K1,s}$, $I_{K1,t}$); plateau potassium currents ($I_{Kp,s}$, $I_{Kp,t}$); sodium-calcium exchange currents ($I_{NaCa,s}$, $I_{NaCa,t}$) and sodium-potassium pump currents ($I_{NaK,s}$, $I_{NaK,t}$).*

Concentration changes in sarcoplasmic reticulum, cytoplasm and TAT-system accompanying the electrical responses (Fig. 4) are shown in Fig. 5. As follows from the figure, the increasing magnitude of Ca_i-transients (Ca_i) reflects the increasing load of Ca-ions in uptake (SR_{up}) and release (SR_{rel}) compartment of sarcoplasmic reticulum at *1 Hz* stimulation frequency. The intracellular Na and K concentrations also tend to reach a new level that is characteristic for this rate. **The most significant feature of the present model is its ability to simulate the dynamic changes of ion concentrations in TAT-system (Ca_t, K_t, Na_t) that are supposed to be involved in processes underlying cellular arrhythmogenesis.**

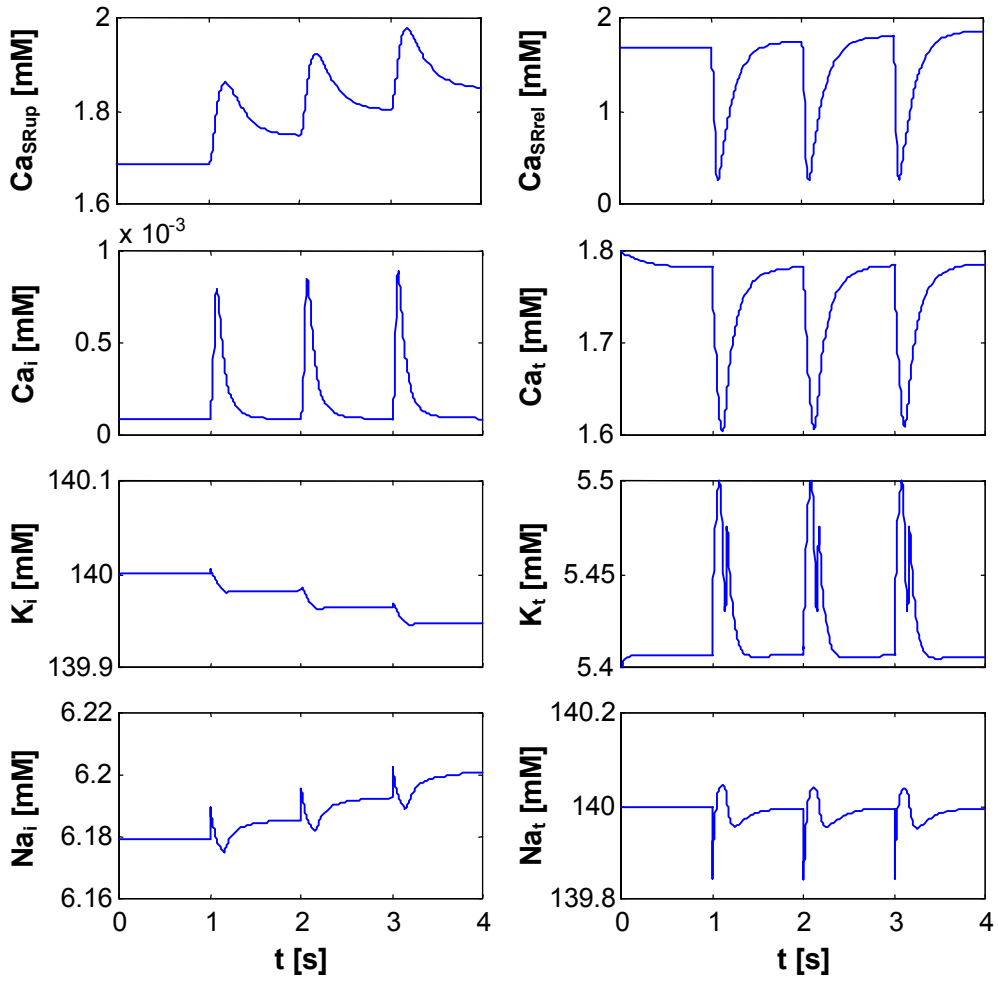

Figure 5. *Changes of ion concentrations in uptake (SRup) and release (SRrel) compartment of sarcoplasmic reticulum, in myoplasm (Ca_i, K_i, Na_i) and in TAT-system (Ca_t, K_t, Na_t) accompanying the excitations in Fig. 4.*

4. CONCLUSION

The described model represents a thoroughly refined version of our former tentative model [27] of ventricular cell electrical activity. It is based on recent available experimental data from isolated guinea pig cardiomyocytes. Besides electrical properties of surface and tubular membrane, the model can also simulate dynamic changes of ion concentrations in myoplasm and in TAT-system. These new features of the model make it possible to simulate the effect of TAT-system on cellular electrical activity as well as to explore quantitatively the role of transient accumulation and depletion of tubular ions in ventricular arrhythmogenesis.

5. ACKNOWLEDGMENTS

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