Post-Repolarization Refractoriness in Human Ventricular Cardiac Cells

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Abstract

Computer simulations have been used to study the mechanisms of postrepolarization refractoriness in cardiac cells under ischemic conditions at the cellular level. The ten Tusscher model of the cardiac action potential has been used with the formulation of the ATP-sensitive K^+ current by Ferrero et al being adopted. Cells were subjected to hyperkalemia, hypoxic and acidic conditions. The results show that the three components of ischemia decrease the action potential duration (APD) as well as the conduction velocity, while effective refractory period (ERP) depicts a non-monotonic behavior. Under hyperkalemic conditions, no supernormal conduction is observed near physiologic values, and conduction relies on $I_{Ca(L)}$ for $[K^+]_o > 11$ mmol/L. Under hypoxic conditions the trend observed in hyperkalemia are maintained but conduction blocking is obtained at a $[K^+]_o$ concentration of 10 mmol/L. This condition minimally affects the conduction velocity of the hyperkalemic tissue. Acidosis gradually increases the difference between ERP and APD for reductions above the 60%, with conduction blocking occuring at 90%.

1. Introduction

During ischemia, the delivery of substrates, primarily oxygen, to the myocardium stops, resulting in a progressive deterioration of the electric activity in the injured region and subsequently to a loss of function and ultimately pump failure [1]. These metabolic changes are mainly hypoxia, increased concentrations of the extracellular K^+ (hyperkalemia), increased concentrations of intracellular Na^+, and Ca^{2+}, decreased concentration of extracellular Na^+, decrease of intracellular ATP, and acidosis[2, 3].

In normal myocardial cells, recovery of electrical excitability coincides in time with the end of the action potential [1]. Therefore, when membrane potential returns to the resting value, the cell might be excited again if stimulated with a suprathreshold stimulus. Under pathologic conditions (i.e., ischemia), the recovery of excitability may outlast repolarization, a phenomenon known as postrepolarization refractoriness. Hyperkalemia causes a depolarization of the mycardium at rest, slowing conduction, depressing excitability and increasing the recovery time of the h gates of sodium channels. Another metabolic abnormality caused by ischemia is hypoxia, which causes a reduction in the ATP/ADP ratio that can slow calcium pumps and exchangers [4],increasing intracellular sodium and calcium concentrations as well as activating ATP sensitive potasium chanels (I_{KATP} channels) [2]. Acidosis reduces the availability of sodium and calcium channels, reducing excitability and slowing conduction. These metabolic abnormalities cause inhomogeneities of resting potential and dispersion in recovery of excitability in the tissue. In addition, these inhomogeneities provide settings for unidirectional blocks which can become the substrate for arrhythmias. Even though it is possible to reproduce unidirectional blocks experimentally, reproducing unidirectional blocks during acute ischemia are difficult.

The aim of this work was to study the effects of hyperkalemia, anoxia and acidosis on recovery of excitability and conduction mechanisms on recent human cardiac action potential models[5, 6] using computer simulations. The APD, ERP as well as conduction velocity were measured and compared for the different cell types subjected to different levels of hyperkalemia, hypoxia and acidosis using one dimensional simulations.

2. Methods

A modified version of the ten Tusscher cardiac action potential model [5, 6] was used in the simulations. The model describes the principal ionic currents through the cardiac cell membrane with high degree of electophysiological detail for the three types of cardiac cells. The basic model was completed with the formulation of the ATP-sensitive K^+ current (I_{KATP}) described by Ferrero et al [7]. Heterogeneites in the I_{KATP} current were not considered in this study.

Action potential generation and propagation in one dimension was described by using the following parabolic

reaction diffusion equation

$$C_m \frac{\partial V}{\partial t} = -(I_{ion} + I_{stim}) + \frac{\partial}{\partial x}\left(\sigma \frac{\partial V}{\partial x}\right), \quad (1)$$

where C_m is the membrane capacitance, V is the transmembrane potential, I_{ion} is the sum of all transmembrane ionic currents, I_{stim} is the externally applied transmembrane current, and σ is de conductivity of the tissue. A multicellular fiber 4 cm long was considered in the numerical study. For the simulations, the operator splitting technique was implemented. Forward Euler integration with a time step $\Delta t = 0.02$ ms was used for integrating the single cell equation. Implicit Euler with a time step $\Delta t = 0.02$ ms and with a space discretization of $\Delta x = 0.01$ cm was used to integrate the parabolic equation. The conductivity of the tissue was set to $\sigma = 0.0013\ cm^2/s$ which yields a conduction velocity of 71 cm/s under normal conditions.

Hyperkalemic conditions were simulated by increasing the extracelular K^+ concentration ($[K^+]_o$) from 4.0 mmol/L to different values depending on the degree of hyperkalemia. Hypoxia was simulated by activating I_{KATP} to 0.55% of its maximum activation value which produced a 50% reduction of the APD, corresponding to 10 minutes of ischemia [8]. Acidosis was simulated by varying the maximum conductances of I_{Na} and $I_{Ca(L)}$ over a wide range, depending on the severity of acidosis[9]. No shifting in the potential of the I_{Na} kinetics was considered in this study.

The stimulation protocol consisted on a train of ten basic stimulations (S1) with a cyclic length of 1s following an extrasystole stimulus (S2) in 1 millisecond steps in order to determine ERP. The APD was measured at the tenth stimulus, and was defined as 90% of depolarization time and was taken as the average of three cells located at the 50%, 75% and 95% of the length of the strang. The ERP was defined as the shortest S1S2 interval such that the S2 propagated successfully through the fiber. The basic and test stimuli consisted on rectangular pulses 3ms in duration and 1.5 times the diastolic threshold in amplitude.

Diastolic threshold was determined as follows. For a given $[K^+]_o$ value, the model was first stabilized until when the product of the gates $h \cdot j$ reached the 99% of the steady state value $h_\infty \cdot j_\infty$. Once the model had reached steady-state conditions, an stimulation current was applied at the left end of the cable. Diastolic threshold was defined as the minimum stimulation current requiered in order to propagate an action potential in the cable.

The safety factor (SF) for conduction [10] was also determined along the multicellular cable in the limit case before block occurs or when a highly depressed conduction occured. The SF for conduction was calculated according to the formula proposed by Romero et al. [11].

3. Results

Figure 1 shows the computed conduction velocity for the condition of elevated $[K^+]_o$, elevated $[K^+]_o$ and anoxia (Figure 1a), and acidosis (Figure 1b). The figure demonstrates that the three components of ischemia decrease the conduction velocity. Elevated concentrations of $[K^+]_o$ has the greatest influence on reducing conduction. However, the model does not show "supernormal conduction" near physiologic values as has been observed in other cardiac cell models [9]. The results also show that anoxia does not very much affect the conduction velocity under hyperkalemic conditions, but blocking occurs at lower $[K^+]_o$ values ($[K^+]_o > 10$ mmol/L). Under hyperkalemic conditions, conduction block occurred at $[K^+]_o > 15$ mmol/L. Acidosis monotonically decreases conduction. At a 60% reduction of I_{Na} and $I_{Ca(L)}$, conduction velocity decreases 28.7% from 71.3 cm/s to 50.9 cm/s, causing conduction block at 90% reduction of both I_{Na} and $I_{Ca(L)}$.

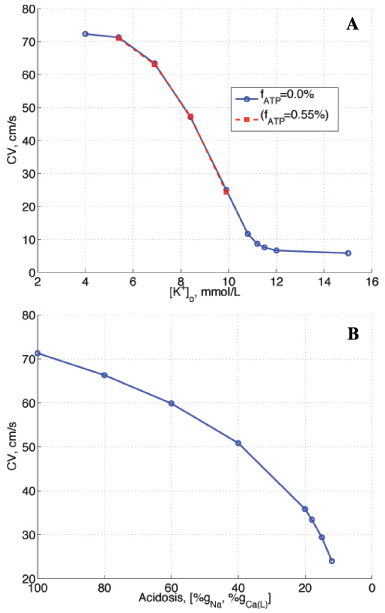

Figure 1. Effect of ischemic conditions on conduction velocity. The three conditions of ischemia were applied with different levels of severity, except for anoxia where only one condition was considered for different levels of hyperkalemia. No supernormal conduction is observed under hyperkalemic conditions. While hyperkalemia and acidosis very much change the conduction velocity, anoxia seems to have no influence on conduction velocity under hyperkalemic conditions.

Figure 2 shows the results for the APD and ERP for the epicardial cell only (a similar behavior was observed for

endocardial and midyocardial cells). Figure 2a shows the APD of the last basic beat and ERP as hyperkalemia increases. Solid lines correspond to conditions of increasing hyperkalemia only ($I_{KATP} = 0$), while dashed lines correspond to conditions of hyperkalemia + hypoxia. When considering hypoxia, the fraction of open I_{KATP} channels, f_{ATP}, was set to 0.55% which causes a 50% reduction in the APD. Figure 2b shows the APD and ERP under acidic conditions.

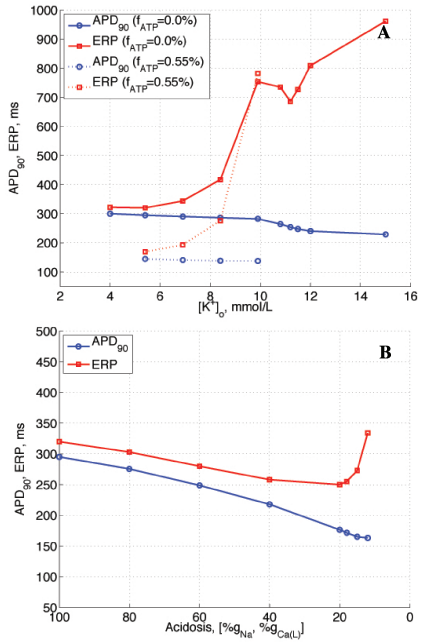

Figure 2. a) Action potential duration (APD) of the last basic beat and effective refractory period (ERP) for different $[K^+]_o$ values and different cell types. Solid lines correspond to normoxia and dashed lines correspond to hypoxia. b) Action potential duration (APD) of the last basic beat and effective refractory period (ERP) for different acidic conditions .

Results demonstrate that all components of ischemia cause postrepolarization refractoriness. For hyperkalemic and hyperkalemic+hypoxic conditions, the difference between ERP and ADP remains almost constant for $[K^+]_o \leq 7$ mmol/L, with this difference increasing rapidly for $[K^+]_o > 7$ mmol/L. For values of $[K^+]_o < 8.4$ mmol/L, the absolute difference between ERP and APD is not significantly different in hyperkalemia and hyperkalemia+hypoxia. However, for $[K^+]_o$ close to 10.0 mmol/L, the ERP value for hyperkalemia+hypoxia overtakes the value in hyperkalemia, with total block occurring in the hypoxic tissue for $[K^+]_o > 10$ mmol/L. For the hyperkalemic tissue, a highly nonlinear behavior is observed for $[K^+]_o > 10$ mmol/L. For acidic conditions, the results show an important influence of acidosis on ERP for reductions in I_{Na} and $I_{Ca(L)}$ above 60%, from which the absolute difference between ERP and APD increases monotonically until reaching the blocking value of 90% reduction.

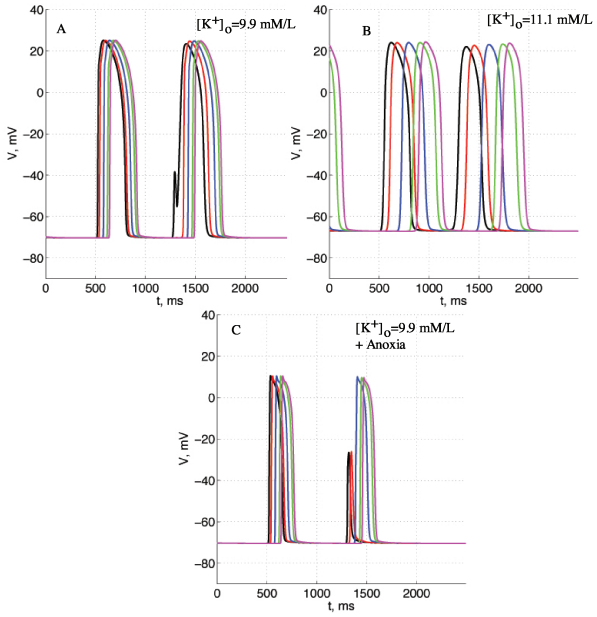

Figure 3. Action potentials under different hyperkalemic+hipoxic conditions. a) $[K^+]_o = 9.99$ mM/L and no hypoxia, b) $[K^+]_o = 11.1$ mM/L and no hypoxia, c) $[K^+]_o = 9.99$ mM/L and hypoxia. The first set of action potentials correspond to the last basic beat, and the second set correspond to the first stimulus for which conduction was observed.

Figure 3 depicts the propagation of action the action potential under different hyperkalemic+hipoxic conditions. Panel A shows the propagation of the action potential for $[K^+]_o = 9.9$ mmol/L, panel B for $[K^+]_o = 11.1$ mmol/L, and panel C for for $[K^+]_o = 9.9$ mmol/L, + hypoxia. The first set correspond to the last basic beat, and the second corresponds to the first stimulus for which conduction was observed. The results show that for $[K^+]_o = 9.9$ the conductions relies on depressed I_{Na}, with a clear interplay of rectifying potassium currents and calcium current at the end of the upstroke which gives a slight biphasic aspect to the action potential. For $[K^+]_o = 11.1$ slow action potentials are obtained. Total suppression of $I_{Ca(L)}$ under this conditions provoked conduction block demonstrating that the action potential and conduction relied on $I_{Ca(L)}$ under severe hyperkalemic conditions. For the case of hyperkalemia + hypoxia, a transition in the conducted action potential is observed. A very depressed and slowing conducting response evolves to a true action potential with conduction relying on depressed I_{Na}. Computations have

shown a safety factor of 1 along the ischemic fiber even for the highly depress action potential of panel C.

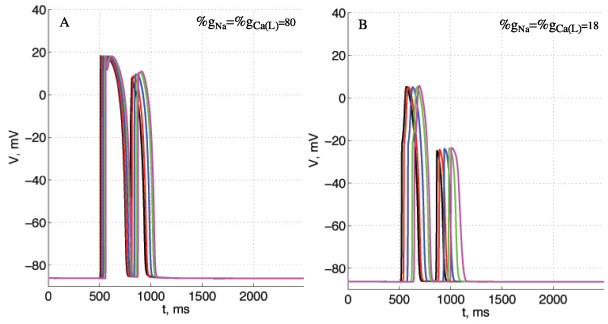

Figure 4. Action potentials under mild and extreme acidosis. a) mild acidosis $\%g_{Na}, \%g_{Ca(L)} = 80$, b) extreme acidosis $\%g_{Na}, \%g_{Ca(L)} = 18$. The first set of action potentials correspond to the last basic beat, and the second set correspond to the first stimulus for which conduction was observed.

Figure 4 shows the propagated action potential under two acid conditions. In both cases conduction appears to rely in I_{Na}, with an important reduction in the maximum of the action potential developed in the fiber. For levels of acidosis close to block, a biphasic action potential upstroke is observed with a very depressed action potential propagating at the ERP. As with hyperkalemia+hypoxia, in this case computations gave a safety factor of 1 along the ischemic fiber.

4. Discussion and conclusions

In this work, computer simulations have been performed to investigate the influence of hyperkalemia, hypoxia, and acidosis on conduction and postrepolarization refractoriness on a human cardiac action potential model. The results show that the absolute difference between APD and ERP increases in hyperkalemia and hyperkalemia+hypoxia, while for acidic conditions an increment in the difference is only observed for reductions above 60% in I_{Na} and $I_{Ca(L)}$. For severe hyperkalemia (These results are in agreement with reported experimental studies and simulations on guinea-pig [9, 10, 12]. Our results also show that the model does not exhibit "supernormal conduction" near physiological $[K^+]_o$ concentrations and that, for severe hyperkalemia, conduction and action potential are supported by $I_{Ca(L)}$ resulting in a lower conduction velocity and a reduction in the ERP. For mild hyperkalemia, postrepolarization refractoriness is due to the effect of cell depolarization on the recovery kinetics of I_{Na}. However, for severe hyperkalemia, postrepolarization refractoriness responds to $I_{Ca(L)}$ kinetics and the slow velocity of the depolarization front. Acidosis reduces conduction velocity and the difference between ERP and APD increases monotonically for reductions in I_{Na} and $I_{Ca(L)}$ above 60% until reaching the blocking value of 90% reduction.

Acknowledgements

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