Afterdepolarizations Promote the Transition From Ventricular Tachycardia to Fibrillation in a Three-Dimensional Model of Cardiac Tissue

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Recent experimental results regarding the action potential duration restitution curve have explained the transition from ventricular tachycardia (VT) to fibrillation (VF) in terms of spiral wave (SW) meandering and breakup. However, it remains unclear whether VF always has a steep restitution curve. The present study was designed to test the hypothesis that afterdepolarizations occur at excitable gaps during VF and affect the SW dynamics, even if the restitution curve is gentle. Homogeneous and isotropic 3-dimensional tissue was simulated with a LRD model. Because of the gentle restitution curve, it was not expected that SW instabilities would occur in this condition. In the tissue, a stationary SW reentry was initially observed; however, afterdepolarizations erupted from the excitable gap near the SW tip, and the SW then meandered widely. Following that, afterdepolarizations erupted far from the SW tip, resulting in SW breakup. In this manner, the wave dynamics degenerated into a chaotic state within a few seconds. Furthermore, not only triggered activity but also subthreshold afterdepolarizations were found to cause SW instabilities. These results suggest that afterdepolarizations may play an important role in the transition to VF and that the mechanism is independent of restitution properties. (*Circ J* 2002; 66: 505–510)

Key Words: Afterdepolarizations; Restitution; Spiral wave; Ventricular fibrillation; Ventricular tachycardia

Recent experimental studies have shown that the spiral or scroll wave (SW) is one of the important mechanisms of ventricular tachyarrhythmias.^{1,2} Clinical observations have suggested that ventricular fibrillation (VF) is usually preceded by ventricular tachycardia (VT),³ and some experiments and simulation studies have suggested that the mechanism of the transition from VT to VF is based on SW meandering^{1,2} and/or breakup.^{4–7} The Restitution Hypothesis^{4,6,7} has explained the mechanism of the SW breakup caused by the interaction between the wavefront and the wavetail: A steep slope (>1) of the action potential duration (APD) restitution curve results in spontaneous SW breakup, and a gentle slope (<1) results in SW stability.

Afterdepolarizations are defined as oscillations in the myocardial cell membrane potential that follow the upstroke of an action potential. In 1962, Reiter⁸ reported that digitalis administration or high calcium concentration produced a series of spontaneous afterdepolarizations (aftercontractions) in guinea pig papillary muscles, and that these afterdepolarizations were induced by a greater rate of stimulation. Early and delayed afterdepolarizations (EADs

and DADs, respectively) induce triggered activity (TA)⁹ that can result in the induction of cardiac arrhythmias.^{10–15} The EADs are classified into phase-2 and phase-3 EADs. The phase-2 EADs can be induced experimentally by various conditions, even in the absence of calcium overload. On the other hand, according to the present view, the DADs and phase-3 EADs are associated with fast rates and calcium overload conditions, and the phase-3 EADs are considered to share a common mechanism with DADs;^{16,17} that is, calcium ions are spontaneously released (calcium-induced-calcium-release) from the overloaded sarcoplasmic reticulum (SR)^{18,19} and this generates an intracellular calcium transient that activates the inward current of sodium-calcium exchanger (INaCa) and the nonspecific calcium-activated current (Ins(Ca)) to depolarize the myocardial cell membrane.¹⁶ In addition, the enhancement of inward INaCa and that of inward Ins(Ca) in response to calcium overload, and their contributions to membrane depolarization, are comparable.

However, there are a number of points regarding the mechanisms of the transition from VT to VF that have not been clarified, and it remains unclear whether VF always has a steep APD restitution curve. Moreover, it is unknown whether the DADs and phase-3 EADs occur while a SW rotates. Accordingly, we hypothesized that these afterdepolarizations occur at excitable gaps during SW reentry and, even if the maximum slope of the APD restitution curve is gentle (<1), the afterdepolarizations will affect the SW dynamics, such as meandering and wave breakup. To confirm this hypothesis, we studied the behavior of SW reentry in 3-dimensional (3-D) computer simulations using a realistic mammalian ventricular action potential model.

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Methods

Simulated Myocardial Tissue

We used the LRd (Luo-Rudy-2) ventricular action potential model^{16,20} without any modifications as the myocardial unit in this study. The validation of the LRd model has been addressed in previous reports.^{16,20} The model includes the following currents: fast sodium (INa), L-type calcium (ICaL), time-dependent potassium (IK), time-independent potassium (IK1), plateau potassium (IKp), sodium-calcium exchanger (INaCa), sodium-potassium pump (INaK), non-specific calcium-activated (Ins(Ca)), sarcolemmal calcium pump (Ip(Ca)), calcium background (ICa,b), sodium background (INa,b), and the time-independent, purely voltage-dependent current (Iv). Furthermore, this model incorporates calcium release (Irel) and uptake (Iup) by the SR, the translocation of calcium between 2 compartments in the SR (the network SR and the junctional SR²¹) (Itr), and the buffering processes in the SR and the myoplasm; therefore, it is possible to simulate the behavior of cells under calcium overload conditions, and the related arrhythmogenic activity, such as EADs, DADs, and TA, of the myocardial cells.

Fig 1. The standard action potential duration (APD) restitution curve (upper solid curved line) and dynamic APD restitution curve (lower solid curved line with black dots). The preceding diastolic interval (DI) is plotted along the horizontal axis, and the APD on the vertical axis. The dotted oblique line represents slope = 1. The maximum slopes of those restitution curves were below 1.

Next, we composed a homogeneous and isotropic 3-D myocardial tissue model, 6.0×6.0×1.0 cm (4,500,000 units). The values of the parameters used to establish the electrical conductivity of the tissue were: the spatial discretization step in all directions was 0.02 cm, the time discretization step was 0.01 ms, the membrane capacitance was 1.0 μF/cm², the conductivity between the myocardial units was 1.0 mS/cm, and the surface-to-volume ratio was 1,800/cm. The conduction velocity of a planar wave was calculated as 0.387 m/s. The tissue borders were set to non-flux Neumann boundary conditions. The SWs were generated by the S1-S2 method, where a premature stimulus in the shape of a part of a globe was administered when a refractory tail of a planar wave had just passed through the center of the simulated tissue.

APD Restitution Curve and Simulated ECG

The standard APD restitution curve was obtained by premature stimuli after a pacing protocol performed 8 times at 500 ms. The APD and the restitution curve were measured in one myocardial unit at a simulated temperature of 37°C with a pacing pulse duration of 5 ms and a pacing strength twice the diastolic threshold. The dynamic APD restitution curve was obtained during fibrillatory conduction in the 3-D myocardial tissue model.

The simulated electrocardiograms (ECGs) were calculated from the signals obtained at a unipolar electrode located 3.0 cm above the center of the epicardial (front) tissue surface. The equation used for the simulated ECG was described in our previous simulation study.²²

Computations

All the computations were performed on an NEC SX-4/16 supercomputer (16GFLOPS; NEC, Tokyo, Japan). We used the forward Euler method for the integration of the LRd equations. It took about 50 h on the supercomputer to process a typical simulation of 3,000 ms of real time for the 3-D tissue. The convergence of the results for each simulation period was tested by repeating some simulations with half of the spatial and time discretization steps. We confirmed that decreasing these discretization steps did not significantly affect the simulation results.

Fig 2. (A) Consecutive snapshots of the transmembrane potential maps during spiral wave (SW) reentry in 3-dimensional cardiac tissue with the original LRd model. The excitation waves are displayed as a gray scale according to the potential (see gray scale bar). Asterisk (R1) and dagger (R2) represent the location of units recording some cellular level data (see Fig 4). (B) Consecutive snapshots of the transmembrane potential maps during SW reentry in the same tissue with the LRd model modifying the calcium ion release from the junctional sarcoplasmic reticulum to the myoplasm (Irel). The excitation waves are displayed as a gray scale according to the potential (see gray scale bar). (C) Simulated electrocardiograms (ECGs) from the transmembrane potential maps during the SW reentries. Black solid [Irel(+); original model] and shaded [Irel(-); modified model] lines represent the simulated ECGs of (A) and (B), respectively.

Results

APD Restitution Curve of the LRd Model

Fig 1 shows the APD restitution curve obtained from the LRd model. We found that the maximum slopes of the APD restitution curves were gentle and less than 1. According to the Restitution Hypothesis^{4,6,7} it was not expected that SW meandering and/or breakup would occur under these myocardial conditions.

Simulations in 3-D Tissue

Fig 2A shows consecutive snapshots of the transmembrane potential maps during SW reentry. After the S2 stimulus (275 ms), a stationary SW reentry (500–1,250 ms) was observed. However, the afterdepolarizations (* in Fig 2A) erupted from the excitable gap adjacent to the SW tip, then the SW became nonstationary and began to meander widely in the myocardial tissue (1,300–1,400 ms). Subsequently, afterdepolarizations († in Fig 2A) also erupted from the excitable gap a little further from the SW tip, and part of the wavefront of the SW arm was disturbed by the afterdepolarization area, resulting in the first SW breakup (1,430–1,450 ms). In this manner, the SW meandering and occasional breakup occurred frequently because the afterdepolarizations randomly erupted from the excitable gap in the tissue; therefore, the wave dynamics degenerated into a chaotic state within a few seconds (1,500–3,000 ms).

Fig 3 shows the trajectory of the SW tip on the epicardial (front) tissue surface during the reentrant activation shown in Fig 2A. The SW tip initially rotated around the fixed small circular core (stationary reentry: 500–1,260 ms), but later the SW tip had an asymmetric and random (not the symmetric petal type) trajectory (meandering: from 1,265 ms), even though the simulated myocardial tissue was completely homogeneous and isotropic. Fig 3 also clearly shows the SW tip trajectory during the SW breakup (wave breakup: from 1,430 ms).

To examine the role of DADs and phase-3 EADs during SW reentry, we mathematically blocked calcium ion release from the junctional SR to the myoplasm (Irel), which directly induced the DADs and phase-3 EADs. This modification did not alter the APD restitution curve (not shown).

Fig 3. The trajectory of the spiral wave (SW) tip on the epicardial (front) tissue surface shown in Fig 2A. Large square shows the border of the epicardial tissue surface. Black solid circles represent the location of the SW tip by 5-ms steps, and black solid lines connecting the black solid circles represent the trajectory of the SW tip. Black solid arrows denote the direction of the meandering and drifting of the SW tip. Numbers beside the black solid circles indicate the simulation time (ms).

As shown in Fig 2B, during the whole simulation period (to 3,000 ms) there was a stationary SW reentry with neither meandering nor breakup because of the inhibition of the afterdepolarizations.

The simulated ECGs calculated from the transmembrane potential maps during the SW reentries are shown in Fig 2C. In the Irel(–) (modified) model, the simulated ECG showed rapid regular and uniform QRS morphology during the whole simulation period (to 3,000 ms). On the other hand, in the Irel(+) (original) model, the simulated ECG initially also showed rapid regular and uniform QRS morphology; however, following the first eruption of the afterdepolarizations, they showed an irregular and multiform QRS morphology (1,250–3,000 ms), which may correspond to that shown on a clinical ECG during the transition from VT to VF.

DADs and Phase-3 EADs During SW Reentry

The tracings in Fig 4 show the transmembrane potential (V_m) and the detailed intracellular calcium dynamics

Fig 4. These traces show the transmembrane potential (V_m), the intracellular calcium concentration ($[Ca]_i$), the L-type calcium channel current (I_{CaL}), the sodium-calcium exchanger current (I_{NaCa}), the calcium concentration of the junctional SR ($[Ca]_{JSR}$), and the calcium release from the junctional sarcoplasmic reticulum to the myoplasm (Irel). These data were recorded at R1 and R2 in Fig 2A. Dotted vertical lines represent the timing of the first delayed afterdepolarization (DAD; black solid triangle) and phase-3 early afterdepolarization (phase-3 EAD; black open triangle). The dotted vertical line in each panel represents the time that the first DAD or phase-3 EAD was induced in each myocardial unit. See text for details.

Fig 5. An example of breakthrough wave in the 3-dimensional cardiac tissue during the spiral wave reentry. The excitation waves in the upper panel are displayed as a gray scale as described in the legend for Fig 2, and those in the lower panel in gray represent voltage greater than -25 mV.

during the SW reentry obtained from 2 myocardial units [R1(*) and R2(+)] shown in Fig 2A. Each myocardial unit was rapidly excited during the SW reentry (cycle length was approximately 110 ms), which was sufficient to cause intracellular calcium overload conditions, especially junctional SR calcium overload (>7 mmol/L), and this activated the Irel. This Irel elicited DAD and phase-3 EAD without or with quite a few ICaL and INaCa enhancements at the excitable gap. As shown in the left panel of Fig 4, the first solid triangle represents a partial depolarization elicited by DAD in response to Irel. The first open triangle represents a delay of terminal repolarization of the preceding action potential because of phase-3 EAD. The second to fourth solid triangles represent the full action potentials elicited by DADs in response to Irel. As shown in the right panel of Fig 4, the first open triangle represents the delay of terminal repolarization because of phase-3 EAD. The first to third solid triangles represent the full action potentials elicited by DADs. The second open triangle represents the full action potential elicited by phase-3 EAD. Thus, the DADs and phase-3 EADs mostly induced TA; however, it is noteworthy that these afterdepolarizations contributed as a refractory area that was able to cause a local conduction block, even though these afterdepolarizations remained subthreshold (refer to the 1,430–1,450 ms panels of Fig 1A, and the corresponding time in the R2 panel of Fig 4). When the local conduction block occurred near the SW tip (eg, first DAD at 1,250 ms in the R1 panel) and at the SW arm (eg, first phase-3 EAD at 1,430 ms in the R2 panel), the SW began to meander and to breakup, respectively. In addition, we found that small TA and afterdepolarizations only caused transient wave breakup and the resulting waves immediately merged into one wavefront (not shown). These results suggest that the size of the afterdepolarization area is one of the important factors involved in SW breakup.

Breakthrough Waves During SW Reentry

The mechanisms of breakthrough waves on the myocardial surface during VF remain unclear, although it has been reported that they may derive from intramural reentry resulting from complex geometry;²³ rotational anisotropy;²² Purkinje-muscle junctions;²⁴ and intramyocardial Purkinje fibers.²⁵ In contrast, as mentioned briefly in a previous study,²⁶ our simulations (see upper panel of Fig 5) also suggested that the afterdepolarizations are another important factors in the mechanism of breakthrough waves during VF. For example, the lower panel of Fig 5 (skeleton view)

clearly shows that the breakthrough wave (black open circle) was not the result of an intramural reentry. This afterdepolarization mechanism of breakthrough waves does not depend on the myocardial wall thickness; therefore, we conjecture that the concept of afterdepolarizations as a factor in the mechanism of breakthrough wave can be extended to a possible role of afterdepolarizations during atrial tachycardia and fibrillation.

Discussion

The major findings of the present study are (1) DADs and phase-3 EADs may play important roles in the mechanisms of 3-D SW meandering and breakup; (2) the mechanisms were independent of the APD restitution curve; (3) TA and afterdepolarizations that remained subthreshold caused local conduction block of wavefronts; and (4) local conduction block adjacent to and far from the SW tip resulted in SW meandering and breakup, respectively.

Conditions of DADs and Phase-3 EADs

Afterdepolarizations (DADs and phase-3 EADs) occur under a variety of conditions that cause an increase in the calcium ion concentration of the myoplasm and SR above normal levels; that is, calcium overload; digitalis toxicity with a greater rate of stimulation;^{10,27–29} catecholamines;^{30–32} quinidine;³³ myocardial hypertrophy;³⁴ myocardial infarction³⁵ etc. Because of these apparently ionic mechanisms of afterdepolarizations and calcium overload with reentrant activation or some pathologic abnormalities, it is plausible that DADs and phase-3 EADs may be induced by repetitive excitation during SW reentry and play important roles in the degeneration into a chaotic state.

Afterdepolarization Mechanism is Independent of Restitution Properties

Fig 1 and 2A–C clearly show that the mechanisms of SW meandering and breakup resulting from the DADs and phase-3 EADs were independent of the APD restitution curve. The SW instability can be simply explained by the collision between the wavefront of the SW arm and the refractory area resulting from the afterdepolarizations. In addition, we extended this afterdepolarization mechanism concept to the generation of breakthrough waves that may cause complexity during SW reentry. That is to say, the afterdepolarizations may cause SW instability even if the APD restitution slope is gentle (<1). This is a quite interesting and important viewpoint for the production of new antiarrhythmic agents.

Antiarrhythmic agents for VF should be investigated based on 2 strategies: The prevention of (1) the initiation of VT by restraining the triggering events and/or altering the properties of reentrant circuits, and (2) the transition from VT to VF. It has been generally considered that suppressing the afterdepolarizations causing TA is effective only for the former strategy, but it was recently suggested that flattening the APD restitution curve might be effective for the latter.^{6,7} We also conceded that a flattened APD restitution curve is effective for the latter strategy even in 3-D tissue with rotational anisotropy.²² On the other hand, in the present study we found that restraining afterdepolarizations is indispensable for both strategies, and our results suggest that only altering the restitution properties is insufficient to stabilize the SW reentry. We would like to stress that the reducing afterdepolarizations and TA (as with the use of

agents such as lidocaine³⁶ doxorubicin³⁷ and calcium blocking agent¹²) may be important as well for the latter strategy.

Comparison With Previous Studies

Chudin et al^{38,39} computationally studied the stability of 2-dimensional (2-D) SW reentry using a modified LRd model. They focused on the intracellular calcium dynamics, and found that the instability in the intracellular calcium dynamics during SW reentry amplified INaCa and prolonged APD, leading to electrophysiologic inhomogeneities and SW instability. This finding is generally compatible with our data (Fig 4); however, they did not study either the 3-D effect or the simulated ECG and, moreover, they did not discuss any strategies for the treatment of VF.

More recently, Pollard et al⁴⁰ also simulated SW reentry in 2-D myocardial tissue with the LRd model and focused on, as in our study, not only the intracellular calcium dynamics but also the mechanisms of SW meandering and breakup resulting from the afterdepolarizations. Their findings⁴⁰ show good agreement with ours⁴¹ but they did not discuss the 3-D effect or the simulated ECG, which we have focused on in this study. Furthermore, they did not discuss the mechanism of the afterdepolarizations from a clinical viewpoint.

Study Limitations

There were several important limitations in the present study. Although the LRd model is one of the latest versions of the mammalian ventricular cell model and can simulate dynamic changes in ionic concentrations, it is based mainly on single-cell data from the guinea pig. Therefore, we know that this mathematical model is not completely concordant with the human physiologic myocardial cell, and that the afterdepolarizations do not occur as easily in physiologic conditions. For example, the critical cycle length causing irregular calcium oscillation leading to periodic Irel was greater than 500 ms, but this may not be an actual physiological condition. As another example, Riccio et al^{42,43} reported that the dynamic restitution slope recorded during rapid pacing and during VF was more than 1; however, the dynamic restitution slope with the present mathematical model was less than 1. Moreover, we did not use the updated LRd model⁴⁴ because we did not focus on potassium currents. From a structural point of view, we did not consider the spatial heterogeneity in refractoriness⁴⁵ and constructed a homogeneous model with a monodomain nature instead of a bidomain nature⁴⁶ because of the present limitations in computer performance and the finite amount of memory. Despite these limitations, we speculate that the DADs and phase-3 EADs are important mechanisms in the degeneration into VF. Additional studies are required to ensure that our findings are valid in a variety of myocardial cell models, and further experimental studies will be needed to elucidate the real mechanisms of the transition from VT to VF.

Conclusion

Our simulation studies suggested that the DADs and phase-3 EADs may play important roles in the transition from VT to VF, and that these mechanisms are independent of the APD restitution curve. Our findings are supported by the fact that VF is more likely to occur in conditions where there is intracellular calcium overload causing afterdepo-

larizations.

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