Electrophysiological Mechanism of Enhanced Susceptibility of Hypertrophied Heart to Acquired Torsade de Pointes Arrhythmias

Tridimensional Mapping of Activation and Recovery Patterns

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Background—Cardiac hypertrophy is associated with increased incidence of sudden death and susceptibility to proarrhythmic effects of antiarrhythmic agents. However, the in vivo electrophysiologic mechanism of the arrhythmias has not been investigated in detail.

Methods and Results—Dose-dependent susceptibility to Torsade de Pointes (TdP) by class III drug dofetilide, 3, 10, and 30 μg/kg, was compared in 6 control dogs (C) and in 5 dogs 6 to 8 weeks after induction of complete atrioventricular block (AVB) that resulted in ventricular hypertrophy (H). Tridimensional ventricular activation and repolarization (R) patterns were simultaneously analyzed from unipolar extracellular electrograms, and local R was measured from activation recovery intervals. Both R and transmural dispersion of R (TDR) were significantly greater in dogs with H compared with C. Dofetilide resulted in cycle length–dependent and dose-dependent prolongation of R, which was more marked in left ventricular endocardium/midmyocardium compared with epicardium, resulting in significant increase of TDR. These changes were more accentuated in dogs with H compared with C. All 5 dogs with H developed TdP at a dose of 3 to 10 μg/kg, whereas only 1 of 6 C dogs developed TdP at 30 μg/kg. TdP was initiated by subendocardial focal activity that infringed on TDR, resulting in functional conduction block and reentrant excitation.

Conclusions—Enhanced susceptibility of hypertrophied heart to class III drugs is attributable to baseline increased TDR and greater dose-related accentuation of TDR compared with nonhypertrophied heart. This provides the electrophysiologic substrate for drug-induced TdP. (Circulation. 2002;105:1128-1134.)

Key Words: hypertrophy • torsade de pointes • long-QT syndrome • antiarrhythmia agents

Cardiac hypertrophy is associated with increased incidence of sudden death1 and susceptibility to proarrhythmic effects of antiarrhythmic drugs, especially class III agents2 attributed to the presence of increased dispersion of ventricular repolarization (DR).3 Spatial DR is a major substrate for reentrant ventricular tachyarrhythmias, including torsade de pointes (TdP) in the long-QT syndrome.4 Several studies have questioned whether marked spatial dispersion of ventricular repolarization is detectable in vivo5–7 and whether class III antiarrhythmic agents result in an increased DR in the hypertrophied heart.8

The present study was conducted to investigate if the hypertrophied heart is associated with increased spatial DR in vivo and an increased susceptibility to proarrhythmic effects of class III antiarrhythmic agents. For this purpose, we investigated a known model of ventricular hypertrophy9 in which dogs with chronic atrioventricular (AV) block develop ventricular hypertrophy secondary to chronic volume overload.10 The susceptibility of those dogs to the proarrhythmic effects of dofetilide, a class III antiarrhythmic agent and a selective blocker of I_{Kr},11 was investigated using tridimensional mapping of ventricular activation and repolarization patterns. Preliminary data have been published in abstract form.12

Materials and Methods

Surgical Preparation

The study was approved by the Animal Studies Subcommittee of the local institutional review board and conformed to guiding principles of the Declaration of Helsinki. Experiments were performed on 5 purpose-bred mongrel dogs (mean weight, 8±1.6 kg). Dogs were preanesthetized with sodium thiopental (17.5 mg/kg intravenously) via cannulation of the cephalic vein. Dogs were then intubated and anesthetized with 1.0% to 2.0% isoflurane (vaporized in 100% O₃) via a positive pressure ventilator (F500, Foregger Company, Inc).
Complete atrioventricular block (AVB) was induced by radiofrequency ablation technique. The resulting complete AVB was monitored for 30 minutes. An endocardial pacing electrode was then positioned into the apex of the right ventricle through the jugular vein and connected to a pacemaker (THERA 8966i, Medtronic) placed in a subcutaneous pocket created on the posterior neck. Ventricular pacing at a rate of 40 beats per minute was maintained for 6 to 8 weeks.

Cardiac dimensions were regularly monitored using Agilent Sonos 5500 echocardiograph (Hewlett-Packard). Values for left ventricle (LV) mass index (LV weight/body weight) and fractional area shortening were measured. Ejection fraction was calculated as (LVEDV–LVESV)/LVEDV, where LVEDV is LV end-diastolic volume and LVESV is LV end-systolic volume. The fractional area change (FAC) was calculated as (LVEDA–LVESA)/LVEDA, where EDA is end-diastolic area at the papillary muscle level and ESV is end-systolic area.

Six to 8 weeks after complete AVB, when echocardiographic evidence of significant left ventricular hypertrophy (LVH) was demonstrated, dogs were reanesthetized and catheters were inserted into the femoral vein for administration of fluids and drugs and the femoral artery for monitoring of blood pressure. Electrocardiographic leads I, II, and III and blood pressure (Statham transducer, Gould, Inc.) were continuously monitored on Electronics for Medicine VR12 monitor (PPG Industries). The heart was exposed by midsternal thoracotomy, and a pericardial cradle was created. In the control (C) group (n=6), acute complete AVB was produced 30 minutes before the chest was opened. On completion of the experiment, the animals were euthanatized by electrical induction of ventricular fibrillation and the heart was removed under general anesthesia.

**Data Acquisition and Isochronal Mapping**

Details of the mapping technique, calculation of activation-recovery intervals (ARIs), and construction of activation and repolarization isochronal maps have been previously reported. Sixty-four plunge-needle electrodes, each consisting of 4 to 8 unipolar electrodes with 1-to-2 mm interelectrode distance, were used for tridimensional mapping of the whole ventricle. Right ventricle plunge needles usually had 4 to 6 electrodes spaced 1 mm apart. On the other hand, interventricular septum plunge needles had 8 electrodes spaced 2 mm apart (Figure 1). Unipolar electrograms were acquired using 2- or 3-variable gain 128-channel multiplexed data acquisition systems (DSC 2000, INET Corp), allowing simultaneous recording of up to 384 channels. The timing of selected landmarks in each activation and recovery complex was automatically computed and stored for later analysis. At the end of experiment, the exact position of the electrodes was identified as previously described. It was easier to position the most epicardial electrode within 0.5 to 1 mm of epicardial surface. The position of the most endocardial electrode was more difficult to control; however, in ~90% of sites, it was within 1 mm of endocardial surface.

**Stimulation Protocol**

Programmed ventricular stimulation at cycle lengths (CLs) of 400, 600, 1000, and 1500 ms was performed using a digital programmable stimulator (Medtronic 5325, Medtronic, Inc) delivering square pulses (3-ms duration) through a bipolar plunge electrode placed in the right ventricle outflow tract.

**Drug Administration**

The drug was dissolved in 250 μL of dimethyl sulfoxide (DMSO). Recordings were performed during infusion of cumulative doses of dofetilide 3, 10, and 30 μg/kg. Solution of 50 μL DMSO in 50 mL saline was used as a control vehicle.

**Statistical Analysis**

Where applicable, data were analyzed using repeated-measures ANOVA and unpaired t test (SYSTAT for Windows, version 5.0, SPSS Inc, Chicago, IL). P≤0.05 was considered statistically significant.

**Results**

Eleven dogs were included in the study. In the control group (n=6), LV mass index was 3.86±0.21 g/kg compared with 5.51±0.40 g/kg in the chronic AVB group (n=5) (P<0.01). The fractional area change was 56±3.8% in control group compared with 62.3±7.4% in chronic AVB group (nonsignificant). As in previous studies of this model, there was also significant hypertrophy of the right ventricle (RV).

**Effects of Dofetilide on Ventricular Repolarization**

Dofetilide had no effect on the activation pattern of the ventricular paced rhythm but resulted in both CL-dependent and dose-dependent prolongation of ventricular repolarization, as reflected by lengthening of the ARIs. Figure 2 illustrates 8 transmural unipolar electrograms recorded across the basolateral wall of the left ventricle from a dog with chronic AVB during infusion of dofetilide 10 μg/kg. The electrodes were spaced regularly at 1 mm. The most endocardial electrode (No. 1) was located 0.5 to 1 mm from endocardial surface, whereas the most epicardial electrode (No. 8) was located 0.5 to 1 mm from epicardial surface. Recordings were obtained during ventricular pacing at CLs of 400, 600, 1000, and 1500 ms. Measurements of ARI were made after stable ARI was obtained at each CL. At CL of 400 ms, epicardial electrodes No. 7 and 8 had the shortest ARIs, whereas midmyocardial electrodes No. 4 and 5 had the
longest ARIs. However, the difference in ARIs between electrodes No. 5 and 7 (2 mm apart) was only 11 ms. At CL of 600 ms, there was lengthening of ARIs, but difference in ARI between electrodes No. 5 and 7 increased only to 13 ms. At CL of 1000 ms, there was significant prolongation of all ARIs, but the increase in ARIs was more pronounced at midmyocardial and endocardial sites compared with epicardial sites. The difference of ARIs between electrodes No. 5 and 7 increased to 40 ms. At CL of 1500 ms, ARIs showed additional increase, but there was very slight change in the maximum dispersion of ARIs.

Figure 3 shows composite data of ARI distribution collected from 10 unipolar plunge-needle recordings in the basolateral wall of LV in a 6×10-mm section from the same experiment shown in Figure 2. Recordings were obtained during control (panel A) and during infusion of dofetilide 10 μg/kg (panel B). ARIs were grouped in bins and plotted as a function of electrode location along the epicardial-endocardial axis. The differences between groups were analyzed by ANOVA. Dofetilide 10 μg/kg resulted in 18% to 20% increase of ARIs at epicardial electrodes No. 1 and 2 and 23% to 25% increase at midmyocardial/endocardial electrodes 3 through 8. During both control and dofetilide infusion, there was a bradycardia-dependent increase of ARI, more marked at midmyocardial/endocardial sites compared with epicardial sites, especially after drug infusion. At 400 and 600 ms CL, ARIs were slightly longer in midmyocardial zones, but the difference was not statistically significant. At 1000 ms, ARIs at midmyocardial zones No. 4 and 5 were significantly longer than ARIs at epicardial zones No. 7 and 8 during both control (A) and dofetilide infusion (B) (P<0.01). Although ARIs at endocardial sites No. 1 and 2 were usually shorter than at midmyocardial sites, the differences were not statistically significant. At CL of 1500 ms, ARIs seemed to increase proportionally at all sites so that the maximum dispersion of ARI across LV wall was only slightly higher compared with CL of 1000 ms.

The Table summarizes the dose-dependent changes in ARI in the control acute AVB group and the chronic AVB group with cardiac hypertrophy. Because of the varying extension of M cells toward the epicardial and endocardial surfaces in the LV free wall and interventricular septum and our finding that maximum transmural dispersion of ARI seemed to occur between epicardial and midmyocardial/endocardial sites, we grouped ARIs of sites No. 7 and 8 as Epi and sites No. 1 through 6 as End/Mid. The difference between the shortest and longest ARI across each needle was calculated as transmural dispersion of repolarization (TDR). All data were obtained during pacing at a CL of 1000 ms. The Table shows that at baseline both ARIs and TDR were longer in the chronic AV block group compared with control (P<0.05). In the control group, dofetilide resulted in dose-dependent moderate prolongation of ARIs (7% at 3 μg/kg, 18% at 10 μg/kg, and 20% at 30 μg/kg). TDR showed no increase at 3 μg/kg, increased slightly from 18.5 to 25 ms at 10 μg/kg, and showed no additional increase at 30 μg/kg. On the other hand, in the chronic AV block group, dofetilide resulted in greater dose-dependent increase of ARI (17% at 3 μg/kg, 24% at 10 μg/kg, and 31% at 30 μg/kg). Similarly, the TDR showed significant dose-dependent increases (20% at 3 μg/kg, 37% at 10 μg/kg, and 75% at 30 μg/kg). The differences between the dose-dependent effects of dofetilide in the control
Dofetilide and TdP

Only 1 of 6 control dogs developed TdP after dofetilide at 30 μg/kg. By contrast, in all dogs with chronic AVB, dofetilide resulted in spontaneous multifocal ventricular beats. TdP developed during dofetilide infusion at 3 μg/kg in 2 experiments and during 10 μg/kg in 3 experiments. In experiments in which TdP developed during a lower dofetilide dose, the arrhythmia could also develop during infusion of higher dose of the drug. One to 4 runs of TdP developed in each experiment. Of a total of 14 episodes of TdP, 12 terminated spontaneously, and 2 degenerated into ventricular fibrillation. The initiating 1 or 2 beats of TdP consistently arose as focal activity from a subendocardial site in left or right ventricle. All subsequent beats were the result of reentrant excitation attributable to infringement of a focal activity on the spatial dispersion of repolarization, resulting in functional conduction block and circulating wave fronts. TdP terminated when reentrant excitation terminated. Functional conduction block usually developed in the LV free wall between epicardial and midmyocardial sites or between the interventricular septum and RV but not in RV free wall. These sites corresponded to areas with increased spatial dispersion of ARIs. Because the development of TdP was dependent on the site and coupling interval of the initiating focal beat in relation to the site and extent of zones of spatial DR, there was not always a one-to-one correlation between the development of TdP and the absolute degree of DR. For example, the average DR in the 2 dogs that developed TdP at 3 μg/kg of dofetilide was not significantly different from that in 2 of the 3 other dogs that failed to develop TdP at this dose. However, in the latter group, TdP developed at 10 μg/kg of dofetilide in association with a greater degree of DR.

Figure 4 shows surface ECG recording from a dog with chronic AVB after infusion of 10 μg/kg of dofetilide at CL of 1000 ms (S1). A spontaneous ectopic beat (V1) initiated a 9-beat run of TdP at an average CL of 195 ms. This was followed by a slower run of multifocal ventricular rhythm at average CL of 430 ms.
numbers on the electrograms represent the calculated ARIs in milliseconds. The figure shows that dofetilide resulted in significant differential prolongation of ARIs at contiguous sites. In both needle electrode recordings, ARIs at midmyocardial sites C and D and G and H increased by \( \approx 54\% \) compared with control, whereas ARIs at epicardial sites A and B and E and F increased by \( \approx 28\% \) compared with control. The differential prolongation of ARIs resulted in marked increase of the dispersion of repolarization between contiguous sites B and C from 20 ms during control to 79 ms after dofetilide and between contiguous sites F and G from 15 to 78 ms, respectively.

The dofetilide-induced accentuation of TDR at critical sites provided the electrophysiologic substrate for the development of zones of functional conduction block and reentrant excitation. This is illustrated in Figure 7, which juxtaposes the repolarization map of section No. 4 of the S1 beat that preceded the onset of TdP and the activation map of the V1 beat that initiated TdP from the same experiment shown in Figures 4 through 6. The bottom left panel shows selected...
electrograms of 2 S1 beats before TdP. The bottom right panel shows the same electrograms of the S1 beat that immediately preceded the onset of TdP as well as the first 4 beats of TdP. The figure clearly shows that the functional conduction block induced by the V1 beat between contiguous sites B and C and F and G occurred at those sites with marked spatial dispersion of repolarization as depicted by the crowded R isochrones. All subsequent beats of TdP were attributable to varying circulating wavefronts. The TdP was terminated when the reentrant wavefront was blocked. The following slower run of multifocal ventricular rhythm was due to focal activity arising from different subendocardial sites.

**Discussion**

The present study demonstrates that volume overload hypertrophy in dog is associated with prolonged ventricular R and increased TDR compared with nonhypertrophied heart. Furthermore, the hypertrophied heart is more susceptible to the R-prolonging effects of the class III antiarrhythmic agent dofetilide. Dofetilide resulted in significantly more prolongation of ventricular R of the hypertrophied heart. More importantly, the drug resulted in differentially greater prolongation of R at endomyocardial and midmyocardial regions compared with epicardial regions, resulting in increased dispersion of TDR. The increased TDR at contiguous myocardial sites represented the primary electrophysiologic abnormality in dogs with cardiac hypertrophy and is accentuated by Ikr blockers. The study provided detailed analysis of TDR because of the high resolution of data points along the needle electrodes.

Previous reports have questioned whether significant spatial DR can be demonstrated in vivo and whether class III agents result in increased DR. In view of our present findings, methodological differences may have to be invoked for the differences between these studies and the present one. In studies by Anyukhovsky et al.5,6 ARIs were derived from bipolar electrograms, which may be less accurate than measurements obtained from unipolar electrograms.15 In another study by Bauer et al.7 a limited section of the LV was investigated by tridimensional recordings. In the study by Gillis et al.8 only epicardial MAP recordings were analyzed. The present study leaves little doubt that significant spatial DR can develop between contiguous ventricular sites in the in vivo canine heart and can provide the primary substrate for functional conduction block and reentrant tachyarrhythmias. Dofetilide-induced prolongation of APD and increased DR was bradycardia dependent. However, there seems to be some differences in the extent of CL-dependent prolongation of R and TDR when compared with the canine anthopleurin-A surrogate model of LQT3.4 The CL dependence in the LQT3 model seems more exaggerated compared with the present model. In other words, the longer the CL, the greater the lengthening of R and DR in the LQT3 model. In the present model, there was a gradual increase of R as the CL prolonged, but the major increase in DR occurred between CLs of 600 and 1000 ms. This may be related to differences in response of M cells to agents that delay Na+ inactivation (LQT3) versus those that depress Ikr (LQT2). In the former situation, the enhanced inward slow Na+ current during the plateau of
AP shows less time dependence of inactivation\textsuperscript{16} compared with $I_{Kr}$.

**Clinical Implications**

The present study demonstrates that a high dose of dofetilide results in prolongation of $R$, increased DR, and, uncommonly, TdP in normal heart. On the other hand, the hypertrophied heart is more susceptible to the proarrhythmic consequences of dofetilide at doses that are considered within the clinical range.\textsuperscript{15} Our data provide the electrophysiologic basis for the reported dose-related incidence of dofetilide-induced TdP in patients.\textsuperscript{17} It also justifies the recent recommendations for dose titration and close monitoring of the effects of the drug in the clinical setting.\textsuperscript{18}

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**References**

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