The Relationship between the Repetitive Extrasystole Threshold and the Ventricular Fibrillation Threshold in the Dog

Non-parallel Changes following Pharmacological Intervention

PATRICE JAILLON, INGELA SCHNITTGER, JERRY C. GRIFFIN, AND ROGER A. WINKLE

SUMMARY We studied the relationship between the repetitive (two or more) extrasystole threshold (RET) and ventricular fibrillation threshold (VFT) in 38 pentobarbital anesthetized dogs. A 100-Hz train of 16 4-msec stimuli was delivered to the right ventricle during the T wave of every 12th paced supraventricular beat. Current was increased in 1-mA steps until ventricular fibrillation occurred. Five measurements were made in each dog at 15-minute intervals. In 10.3% of the VFT determinations, VF was not preceded by a repetitive extrasystolic activity. For the remainder regression analysis of the correlation between RET and VFT revealed an r = 0.37 (P< 0.001). The average RET was 54.6% of the VFT (5.4 ± 1.6 vs. 10.2 ± 2.4 mA); however, it ranged from 5% to 98%. Five animals received a 90-minute lidocaine infusion (0.3 mg/kg per min). Although lidocaine significantly increased both RET and VFT as a linear function of log lidocaine plasma concentration (Cp), the mean slope of the VFT vs. log lidocaine Cp regression line was significantly different from that of RET vs. log lidocaine Cp regression line (P < 0.05). Nine dogs infused with the β-blocker acebutolol (0.5 mg/kg in five and 2 mg/kg in four dogs) showed a concentration-dependent increase of VFT without significant increase of RET, and the mean slopes of the response vs. log Cp regression lines were significantly different (P <0.01). We conclude that in the control state the RET may be related to the VFT. However, during pharmacological studies, VFT and RET do not necessarily change in parallel. Circ Res 46:599-605, 1980

ELECTRICAL stimulation of the atria or ventricles in the vulnerable period of the cardiac cycle may induce repetitive depolarizations when the current intensity is less than that causing fibrillation (Brooks et al., 1951; Hoffman et al., 1951). This repetitive extrasystole threshold (RET) has been justified in previous studies as an index of the ventricular vulnerability to fibrillation (Corbalan et al., 1974; Han et al. 1964; Hoffman et al., 1951, 1955a, 1955b; Lown et al., 1973; Matta et al., 1976; Rabinowitz and Lown, 1978; Rabinowitz et al., 1976; Rotenberg et al., 1978). However, the assumption that RET reflects the ventricular vulnerability to fibrillation remains controversial because: (1) the current determining VFT is different from that of RET (Shumway et al., 1957), (2) in about 10% of the experiments VF is not preceded by any ventricular repetitive activity (Matta et al., 1976), (3) under conditions of either coronary occlusion (Logic, 1975) or regional hyperkalemia (Logic, 1973a; Logic, 1973b) the RET and VFT do not change in parallel and VF generally is not preceded by repetitive extrasystoles and, (4) after administration of glucose, insulin, and potassium the VFT continues to increase linearly while the RET reaches a plateau (Obeid et al., 1978). The VFT model has been used widely to evaluate the antifibrillatory properties of antiarrhythmic drugs and the measurement of RET rather than VFT would offer certain advantages in these studies. We therefore examined the relation between RET and VFT in the control state and after administration of the antiarrhythmic drugs, lidocaine and acebutolol.
Methods

Experimental Procedures

Dog Preparation
Thirty-eight mongrel dogs weighing 14–29.5 kg were anesthetized with sodium pentobarbital, 30 mg/kg, iv. The dogs were intubated with a cuffed endotracheal tube and ventilated with humidified room air, using a Harvard pump. A large-bore catheter was inserted into the left femoral artery to monitor the systemic arterial pressure using a Statham P23Db transducer. Mean aortic pressure was derived electronically. Blood samples for arterial blood gas analysis were drawn throughout the experiment and were analyzed using a Corning Model 165 blood gas analyzer. Depth and rate of respiration were adjusted to maintain an arterial P02 greater than 72 mm Hg and pH between 7.35 and 7.44 throughout the experiment. A catheter was placed in the right femoral vein for administration of additional anesthetic, which was given as needed to maintain deep anesthesia throughout the experiment. The right jugular vein was catheterized for drug infusions. The chest was opened by means of a midline sternotomy and the heart was exposed and supported in a pericardial sling. The sinus node was crushed and a bipolar pacemaker electrode was attached to the right atrium. Atrial pacing at a rate of 150 beats/min (cycle length 400 msec) was maintained using one channel of a Grass model S88 stimulator and isolation unit.

RET and VFT Determinations
These thresholds were determined using the method of Moore and Spear (1975), with minor modifications which have been published previously (Schnitger et al., 1978). Briefly, a bipolar fibrillation electrode with circular 1.0-mm platinum poles embedded in acrylic and separated by 10.0 mm was sutured to the right ventricular epicardium. The fibrillating current was delivered by means of specially designed digital timing circuitry and a Grass S88 stimulator with a constant current device. The fibrillating current, configured as a train of 16 rectangular pulses, 4 msec in duration and of 100-Hz frequency, was introduced after every 12th supraventricular beat and increased in 1-mA increments until VF occurred. The two supraventricular paced beats after each fibrillation train were inhibited. The fibrillating train was timed so that it began 50 msec after the QRS complex. In all dogs, the train spanned the entire vulnerable period of the cardiac cycle with the train usually ending approximately 30 msec after the T wave of the electrocardiogram and in all instances before the T wave of the evoked beat. The current delivered to the fibrillation electrode was monitored continuously as the voltage drop across a precision (±1%) 100-Ω resistor which was displayed on a Tektronics storage oscilloscope. The minimal level of current producing two or more extrasystoles was defined as the repetitive extrasystole threshold, and that producing sustained ventricular fibrillation as the ventricular fibrillation threshold. Each threshold determination was recorded on a model SP 700 Ampex tape recorder and subsequently was printed on a Honeywell model 1508 Visicorder to verify the timing of and response to each train. The heart was defibrillated directly within 10 seconds using 25-watt seconds delivered from a Hewlett-Packard model 780 2C defibrillator delivered through 5-cm paddles applied directly to the heart. RET and VFT were measured at 15-minute intervals throughout the study.

Drugs
Acebutolol hydrochloride (1% solution) and lidocaine hydrochloride (2% solution), were expressed in terms of salt weight and were diluted in normal saline solution. They were administered by continuous 90 minute intravenous infusion using a Harvard infusion pump. Blood samples for drug concentrations were obtained via the left femoral arterial catheter each time ventricular fibrillation was induced and were replaced with a like volume (5 ml) of saline solution (0.9% NaCl). Plasma concentrations (Cn) of lidocaine were determined using a gas chromatographic method (Benowitz and Rowland, 1973). Acebutolol concentration was measured using a high pressure liquid chromatographic method (Meffin et al., 1977).

Experimental Protocol

Group 1 (Control State)
Two hundred and four VFT determinations were performed during the control state in 38 dogs, (6.4 determinations per dog), the first determination beginning approximately 1 hour after anesthesia. For each of these 38 animals, the mean, the standard deviation (SD), and the coefficient of variation (CV) were calculated for both the RET and the VFT. These mean values and coefficients of variation were then averaged for the entire group. The correlation coefficient between each value of VFT and RET was calculated and the regression line was determined by the least-squares method.

For the lidocaine and acebutolol infusions (groups 2 and 3), each dog served as its own control. Before drug infusion in each experiment, at least four consecutive control measurements of RET and VFT were obtained. The average of these values was taken as the control RET and VFT for that animal.

Group 2 (Lidocaine Infusion)
Five dogs from the 38 of group 1 were studied. After determination of RET and VFT control values in each dog, lidocaine was administered over 90 minutes at a rate of 0.3 mg/kg per min. VFT and RET were determined every 15 minutes until 200
minutes after the beginning of infusion in four dogs and until 290 minutes in one dog.

**Group 3 (Acebutolol Infusion)**

Nine dogs from the 38 of group 1 were studied in this group. After determination of control RET and VFT values in each dog, acebutolol was administered over 90 minutes at a rate of 2 mg/kg in four dogs and 0.5 mg/kg in five dogs. RET and VFT were determined every 15 minutes during and after infusion until 245 minutes after the beginning of infusion in seven dogs and until 155 and 200 minutes in the remaining two.

**Statistical Analysis**

All results are expressed as mean ± sd. During and after the drug infusions, the coefficient of correlation between each value of RET and VFT was calculated and the regression line was determined by the least-squares method. For each dog in groups 2 and 3, the relationship between RET and VFT and the logarithm of the drug plasma concentration was studied using a log-linear model. The slope of the regression line (least-squares method) was calculated for each dog (Tables 1 and 2) and averaged by the least-squares method. For each dog in groups 2 and 3, the relationship between RET and VFT was determined and the independent group Student's t-test was used to compare mean slopes of the concentration response relationship for RET and VFT. The level of significance was fixed at P < 0.05 (two-tailed). Blood pressures at the beginning and end of control VFT measurements were compared using a two-tailed t-test for matched pairs.

**Results**

**Group 1—Control Group**

Of the 38 dogs in which VFT was determined, two (5.3%) had no repetitive ventricular activity before any of four successive VFT measurements. The mean VFT in these animals was 7.4 ± 0.6 and 6.8 ± 0.7 mA, respectively. In the 36 remaining dogs, VFT was determined 196 times and was preceded by repetitive extrasystoles 183 times. Therefore, 10.3% of a total of 204 VFT determinations were not preceded by repetitive ventricular activity.

**Table 1 The Slope and Intercept of the Relationship between RET and VFT and Log C₀ of Lidocaine for Each Dog**

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>RET—Log C₀</th>
<th>VFT—Log C₀</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Slope</td>
<td>Intercept</td>
</tr>
<tr>
<td>1</td>
<td>9.4</td>
<td>2.9</td>
</tr>
<tr>
<td>2</td>
<td>18.5</td>
<td>−3.5</td>
</tr>
<tr>
<td>3</td>
<td>6.8</td>
<td>0.86</td>
</tr>
<tr>
<td>4</td>
<td>14.3</td>
<td>−2.2</td>
</tr>
<tr>
<td>5</td>
<td>0.55</td>
<td>7.1</td>
</tr>
<tr>
<td>Mean</td>
<td>9.9</td>
<td>1.03</td>
</tr>
<tr>
<td>SD</td>
<td>±6.9</td>
<td>±4.2</td>
</tr>
</tbody>
</table>

**Table 2 The Slope and Intercept of the Relationship between RET and VFT and Log C₀ of Acebutolol for Each Dog**

<table>
<thead>
<tr>
<th>Dog no.</th>
<th>RET—Log C₀</th>
<th>VFT—Log C₀</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Slope</td>
<td>Intercept</td>
</tr>
<tr>
<td>1</td>
<td>−1.91</td>
<td>13.56</td>
</tr>
<tr>
<td>2</td>
<td>0.047</td>
<td>7.05</td>
</tr>
<tr>
<td>3</td>
<td>−1.67</td>
<td>9.42</td>
</tr>
<tr>
<td>4</td>
<td>2.69</td>
<td>−0.68</td>
</tr>
<tr>
<td>5</td>
<td>−0.41</td>
<td>6.67</td>
</tr>
<tr>
<td>6</td>
<td>−2.29</td>
<td>17.44</td>
</tr>
<tr>
<td>7</td>
<td>17.97</td>
<td>25.27</td>
</tr>
<tr>
<td>8</td>
<td>1.52</td>
<td>4.04</td>
</tr>
<tr>
<td>9</td>
<td>0.77</td>
<td>5.42</td>
</tr>
<tr>
<td>Mean</td>
<td>−1.025</td>
<td>9.8</td>
</tr>
<tr>
<td>SD</td>
<td>±3.08</td>
<td>±7.8</td>
</tr>
</tbody>
</table>

Of the 183 RET and VFT measurements made in 36 dogs, the mean RET was 5.4 ± 1.6 mA (range 2.3 to 8.7) and the mean VFT was 10.2 ± 2.4 mA (range 5 to 15.7) with an average CV of 26% and 16%, respectively. This difference in average CV is significant (P < 0.001). The correlation between the RET and VFT values was r = 0.37 (P < 0.001), and the regression line is shown on Figure 1. For the 183 VFT measurements preceded by repetitive extrasystoles, the RET averaged 54.6 ± 12.6% of the VFT. However, as seen on Figure 2, there was a wide range for this RET/VFT ratio from 5 to 98%.

Additionally, there was a wide range of the average CV is 19.4 ± 13% to 78 ± 8%. There also was considerable variability of the ratio in each dog from one VFT determination to another one (Fig. 3). The majority of trains applied at currents higher than the RET (but below the VFT) resulted in multiple ventricular responses. However, it was not uncommon for currents higher than the RET to result in only a single evoked response. In fact, 43% of the 183 repetitive extrasystole thresholds had one or more trains at currents above the RET which resulted in only a single evoked ventricular beat.

Mean arterial pressure remained constant during the period when the control VFT measurements were made and was 111.6 ± 17.1 mm Hg before the initial VFT measurement, and 111.1 ± 16.9 mm Hg before the final control VFT measurement.

**Group 2—Lidocaine Infusion**

Before the beginning of the infusion, repetitive extrasystoles preceded VF 24 times out of 26 VFT measurements. The mean value of RET was 5.3 ± 2 mA and mean VFT was 10.6 ± 2.4 mA. The correlation coefficient between each value of RET and VFT was 0.72 (P < 0.001). During and after lidocaine infusion, of 76 VFT determinations RET was present in 66 (87%) and the correlation coefficient between each value of RET and VFT was 0.41 (P < 0.001).
Both RET and VFT increased with log \( C_p \) lidocaine, and the slopes of the regression lines of individual dogs are listed in Table 1. The mean slopes of both the RET — log \( C_p \) lidocaine regression and the VFT — log \( C_p \) lidocaine regression were significantly different from 0 (\( P < 0.05 \) and \( P < 0.01 \), respectively). Of interest, however, was the fact that there was a nonparallel change in RET and VFT with increasing lidocaine \( C_p \) as indicated by a significant difference between the slopes of the RET and VFT — log \( C_p \) lidocaine regression lines (Fig. 4) (\( P < 0.05 \)).

**Group 3—Acebutolol Infusion**

In the control state, two dogs failed to show any repetitive ventricular activity before VF. In the seven remaining dogs, RET was present during 44 of 46 VFT determinations. The mean RET was 5.1 ± 2.2 mA and the mean VFT was 9.9 ± 2.8 mA. The correlation coefficient between each value of RET and VFT was \( r = 0.58 \) (\( P < 0.001 \)). During and after acebutolol infusion, of 144 VFT determinations, RET was present in 131 cases (91%) and the correlation coefficient between each value of RET and VFT was only \( r = 0.22 \) (\( P < 0.05 \)).

The slopes of the log acebutolol \( C_p \) — VFT and log acebutolol \( C_p \) — RET regression lines for indi-
The ventricular vulnerability to fibrillation has been estimated by measuring the ventricular fibrillation threshold, i.e., the least-intense electrical stimulus applied to the ventricle which induces fibrillation. A variety of electrical stimuli have been introduced to induce ventricular fibrillation, including single stimuli which are moved back and forth across the T wave (Wiggers and Wegria, 1940-41), trains of stimuli which span the T wave (Han, 1969) sequential R/T stimulation with three stimuli (Axelrod et al., 1975), and the application of continuous currents. Early work demonstrated that the ventricle is most vulnerable to ventricular fibrillation at those intervals during the cardiac cycle which show a dip in the excitability-recovery curve (Hoffman et al., 1951). A progressive increase in the intensity of the electrical stimulus results in a progressive increase in the number of multiple ventricular responses preceding the onset of ventricular fibrillation (Matta et al., 1976). The use of the RET as an index of ventricular vulnerability to fibrillation has been proposed because: (1) the susceptibility to repetitive extrasystole (RE) occurs at the same intervals as vulnerability to ventricular fibrillation in the auricular and ventricular excitability curves (Hoffman et al., 1951; Hoffman et al., 1955; Hoffman et al., 1955), (2) the VFT is only slightly greater than the RET (Hoffman et al., 1951; Hoffman et al., 1955; Hoffman et al., 1955) (3) defibrillation shocks are not needed, thus making possible repeated measurements at short time intervals (Han et al., 1964), as well as allowing studies in conscious animals (Lown et al., 1973; Corbalan et al., 1974, and (4) under conditions of stellate ganglion stimulation, vagal stimulation and β blockage with practolol, the two measurements change in parallel (Matta et al., 1976).

The RET initially was used as an index of VFT during studies using single stimuli which were moved back and forth across the T wave. Although Shumway et al. (1957) cautioned that use of the RET would seriously distort the results because several milliamperes often separated the shock provocative of extrasystoles from that capable of initiating ventricular fibrillation, the measurement was used as an index of VFT during both control and intervention situations (Corbalan et al., 1974; Han et al., 1964; Hoffman et al., 1951, 1955a, 1955b; Lown et al., 1973; Matta et al., 1976; Rabinowitz and Lown, 1978; Rabinowitz et al., 1976; Rotenberg et al., 1978). The only major analysis of the RET as an index of VFT was by Matta et al. (1976), who carefully examined the relationship between the two, using VF induced by single stimuli. They defined the RET as the lowest current producing repetitive extrasystoles in two of three determinations. Using this definition, they found the RET to be a stable index of VFT and found that it occurred reproducibly at 66 ± 4% of the VF threshold current. They noted that 9% of their animals (3 of 32) failed to have repetitive extrasystoles before VF and explained this observation in 2 of the 3 by the presence of heart worm infestation. These authors found that the RET and VFT changed in parallel after stellate ganglion and vagal stimulation as well as after pharmacologic β blockade with practolol. They did warn, however, that under other conditions a precise correspondence between RE and VF thresholds may not exist.

The findings of our study suggest that one must be cautious when measuring the RET as an index of VFT. Our study does confirm that, in the control state, the RET is an index of the ventricular fibrillation threshold. However, several observations suggest that it may be less ideal than measurement of the VFT. Although statistically significant, the correlation coefficient RET and VFT was only 0.37 (Fig. 1). As pointed out by Matta et al. (1976), 10% of the animals do not experience repetitive extrasystoles prior to developing ventricular fibrillation. In contrast to the findings of Matta, rather than finding RET as a constant fraction of the VFT, we found considerable variability from one animal to the next and from one measurement to the next in a given animal. The fact that the coefficient of variation of the RET was significantly greater than that for the VFT can in large part be explained by the fact that we used current increment increases.
of 1 mA, and the mean value of RET was a smaller number than that of the VFT. The variability of these measurements and the relationship between them did not seem to be a function of the duration of the study, the mean arterial pressure, or doses of anesthesia. The most important observation of our study, however, was the nonparallel change in RET and VFT observed during two different pharmacological studies, one with lidocaine and one with the β-adrenergic-blocking agent, acebutolol. After lidocaine, the mean slopes of the RET and VFT concentration-response regression lines were significantly different with lidocaine, producing a greater effect on VFT compared to RET. This difference would not have changed the overall conclusion of the experiment if RET had been used instead of VFT, since both measurements increased significantly during and after lidocaine. However, after acebutolol, there was a more serious dissociation of the RET/VFT relationship. VFT increased linearly with the acebutolol plasma concentration, whereas RET did not change significantly. In this situation one would draw different conclusions from the experiment if RET were measured rather than VFT.

When comparing the results of our study to those of Matta et al. (1976) and other published data, it is important to keep in mind differences in experimental technique which could account for different conclusions. Matta et al. used a single electrical pulse scanning the vulnerable period which was delivered to the right ventricular endocardium using a catheter positioned at the apex via a jugular vein. In our study, we used a train of rectangular pulses covering the vulnerable period and delivered to the epicardium of the right ventricle. In addition, our measurements were carried out during atrial drive at a rate of 150/min, and the study of Matta was carried out during ventricular drive at a rate of 215 beats/min. The differences between the two methods may explain the differences found in the control values of the VFT (10.2 ± 2.4 mA in our studies vs. 35.6 ± 1.7 mA in the study of Matta et al.) and may explain some of the other differences in the studies. Another important difference between the two studies is that the study of Matta et al. examined the influence of physiological or pharmacological interventions only at a single point, whereas we examined the relationship between RET and VFT over a wide range of plasma concentrations. Finally, the studies examined the relationship between RET and VFT after different interventions.

Our observation of a non-constant relationship between RET and VFT following pharmacological intervention is a finding that occurs in other situations, using a variety of interventions and experimental techniques. In studies of cat and turtle ventricles, Hoffman et al. (1961) noted that after numerous fibrillation/defibrillation episodes initiated by single stimuli, it often became impossible to obtain extrasystoles during the vulnerable period and that the fibrillation threshold fell to a value below that previously determined as the threshold for extrasystoles. This finding implies a non-parallel decline in the RET and VFT. In a more recent study of the influence of glucose, insulin, and potassium on the vulnerability to ventricular fibrillation in the canine heart, Obeid et al. (1978) found a nonparallel change in the VFT and RET. During the first hour of glucose, insulin, and potassium infusion, the RET and VFT both increased in parallel. Thereafter, however, the RET did not increase further whereas the VFT continued to rise. This study also used a single stimulus to initiate the ventricular fibrillation. Logic (1973a, 1973b, 1975) found that, in the control state, the current for initiating multiple ventricular responses was always less than that for initiating ventricular fibrillation. However, after coronary ligation (Logic, 1975) and regional hyperkalemia (Logic, 1973a, 1973b), it was most common for the RET and VFT to be identical (i.e., the first multiple ventricular extrasystoles degenerated into ventricular fibrillation). Finally, Rosati et al. (1966) commented on the interesting finding that in one animal given propranolol, 4 mg/kg, and one given pronethalol, 4 mg/kg, there was an increase of the VFT but no change in the multiple response threshold. This is similar to the results in our acebutolol group.

Although previous investigators have stated that the RE and VF phenomena share a common electrophysiological basis (Matta et al., 1976), the dissociation of these two phenomena after pharmacological or other interventions suggest that they may reflect different phenomena. The events causing the initial few beats after electrical stimulation may be different from those permitting a sustained episode of ventricular fibrillation. An antiarrhythmic drug or other intervention might change these to differing degrees and the net result being a nonparallel change in the RET and VFT.

The nonparallel changes in RET and VFT in our study have important implications for the use of these measurements in intervention studies. If the RET is to be used as an index of ventricular vulnerability to fibrillation, a constant relationship between RET and VFT must be documented over all plasma concentrations or experimental conditions to be studied. Unless a constant relationship is documented, the RET should not be used in place of the VFT.

References
ventricular arrhythmias during myocardial infarction in the conscious dog. Am J Cardiol 34: 692-696
Logic JR (1973a) Experimental effects of anti-arrhythmic drugs on ventricular fibrillation thresholds altered by regional hyperkalemia. Arch Int Pharmacodyn 204: 268-274
The relationship between the repetitive extrasystole threshold and the ventricular fibrillation threshold in the dog. Non-parallel changes following pharmacological intervention.

P Jaillon, I Schnittger, J C Griffin and R A Winkle

_Circ Res._ 1980;46:599-605
doi: 10.1161/01.RES.46.5.599

_Circulation Research_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1980 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/46/5/599.citation

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in _Circulation Research_ can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to _Circulation Research_ is online at:
http://circres.ahajournals.org/subscriptions/